

(19) World Intellectual Property Organization
International Bureau(43) International Publication Date
16 August 2007 (16.08.2007)

PCT

(10) International Publication Number
WO 2007/092638 A1

(51) International Patent Classification:

C07D 271/06 (2006.01) **A61P 3/00** (2006.01)
C07D 333/16 (2006.01) **A61P 19/00** (2006.01)
C07D 419/04 (2006.01) **A61K 31/133** (2006.01)
C07F 9/09 (2006.01) **A61K 31/381** (2006.01)
C07C 215/08 (2006.01) **A61K 31/4245** (2006.01)
C07C 215/10 (2006.01) **A61K 31/661** (2006.01)
A61P 29/00 (2006.01) **A61K 31/6615** (2006.01)

(21) International Application Number:

PCT/US2007/003645

(22) International Filing Date: 9 February 2007 (09.02.2007)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:

60/855,960 1 November 2006 (01.11.2006) US
 60/860,694 21 November 2006 (21.11.2006) US
 60/771,789 9 February 2006 (09.02.2006) US

(71) Applicant (for all designated States except US): UNIVERSITY OF VIRGINIA PATENT FOUNDATION

[US/US]; 250 West Main Street, Suite 250, Charlottesville, VA 29902 (US).

(72) Inventors; and

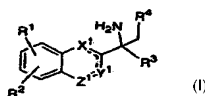
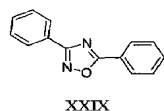
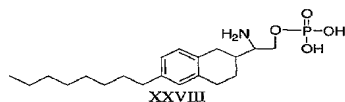
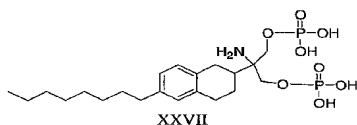
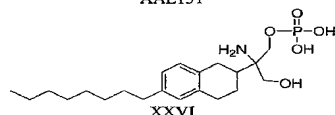
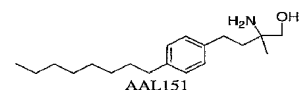
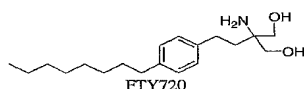
(75) Inventors/Applicants (for US only): **LYNCH, Kevin, R.**; 1632 Meadowbrook Heights Road, Charlottesville, VA 22901 (US). **MACDONALD, Timothy, L.** [US/US]; 200 Douglas Avenue, Unit 3B, Charlottesville, VA 22902 (US).(74) Agent: **PROUT, William, F.**; IPLM Group, P.A., Post Office Box 18455, Minneapolis, MN 55418 (US).

(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

[Continued on next page]

(54) Title: BICYCLIC SPHINGOSINE 1-PHOSPHATE ANALOGS

(57) Abstract: Compounds that have agonist activity at one or more of the S1P receptors are provided. The compounds are sphingosine analogs that, after phosphorylation, can behave as agonists at S1P receptors.



WO 2007/092638 A1



(84) **Designated States** (*unless otherwise indicated, for every kind of regional protection available*): ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

— *with international search report*

— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

BICYCLIC SPHINGOSINE 1-PHOSPHATE ANALOGS

Cross-Reference to Related Applications

[0001] This application claims priority to Provisional Application Serial Nos. 60/771,789, filed February 9, 2006, 60/855,960, filed November 1, 2006 and 60/860,694, filed November 21, 2006, the disclosures of which are incorporated by reference in their entirety.

US Government Rights

[0002] This invention was made with United States Government support under Grant No. RO1 GM 067958 awarded by the National Institutes of Health. The United States Government may have certain rights in the invention.

Background of the Invention

[0003] Sphingosine 1-phosphate (S1P) is a lysophospholipid mediator that evokes a variety of cellular responses by stimulation of five members of the endothelial cell differentiation gene (EDG) receptor family. The EDG receptors are G-protein coupled receptors (GPCRs) and on stimulation propagate second messenger signals via activation of heterotrimeric G-protein alpha (G_α) subunits and beta-gamma ($G_{\beta\gamma}$) dimers. Ultimately, this S1P-driven signaling results in cell survival, increased cell migration and, often, mitogenesis. The recent development of agonists targeting S1P receptors has provided insight regarding the role of this signaling system in physiologic homeostasis. For example, the immunomodulator, FTY720 (2-amino-2-[2-(4-octylphenyl) ethyl] propane 1,3-diol), that following phosphorylation, is an agonist at 4 of 5 S1P receptors, revealed that enhancing S1P tone influences lymphocyte trafficking. Further, S1P type 1 receptor (S1P₁) antagonists cause leakage of the lung capillary endothelium, which suggests that S1P may be involved in maintaining the integrity of the endothelial barrier in some tissue beds.

[0004] Sphingosine 1-phosphate (S1P) is a lysophospholipid mediator that evokes a variety of cellular responses by stimulation of five members of the endothelial cell differentiation gene (EDG) receptor family.

[0005] Sphingosine-1-phosphate (S1P) has been demonstrated to induce many cellular processes, including those that result in platelet aggregation, cell proliferation, cell morphology, tumor-cell invasion, endothelial cell chemotaxis and angiogenesis. For these reasons, S1P receptors are good targets for therapeutic applications such as wound healing and tumor growth inhibition.

[0006] Sphingosine-1-phosphate signals cells in part via a set of G protein-coupled receptors named S1P₁, S1P₂, S1P₃, S1P₄, and S1P₅ (formerly EDG1, EDG5, EDG3, EDG6 and EDG8). The EDG receptors are G-protein coupled receptors (GPCRs) and on stimulation propagate second messenger signals via activation of heterotrimeric G-protein alpha (G_α) subunits and beta-gamma (G_{βγ}) dimers. These receptors share 50-55% amino acid sequence identity and cluster with three other receptors (LPA₁, LPA₂, and LPA₃ (formerly EDG2, EDG4 and EDG7) for the structurally related lysophosphatidic acid (LPA).

[0007] A conformational shift is induced in the G-Protein Coupled Receptor (GPCR) when the ligand binds to that receptor, causing GDP to be replaced by GTP on the α-subunit of the associated G-proteins and subsequent release of the G-proteins into the cytoplasm. The α-subunit then dissociates from the βγ-subunit and each subunit can then associate with effector proteins, which activate second messengers leading to a cellular response. Eventually the GTP on the G-proteins is hydrolyzed to GDP and the subunits of the G-proteins reassociate with each other and then with the receptor. Amplification plays a major role in the general GPCR pathway. The binding of one ligand to one receptor leads to the activation of many G-proteins, each capable of associating with many effector proteins leading to an amplified cellular response.

[0008] S1P receptors make good drug targets because individual receptors are both tissue and response specific. Tissue specificity of the S1P receptors is desirable because development of an agonist or antagonist selective for one receptor localizes the cellular response to tissues containing that receptor, limiting unwanted side effects. Response specificity of the S1P receptors is also of importance because it allows for the development of agonists or antagonists that initiate or suppress certain cellular responses without affecting other responses. For example, the response specificity of the S1P

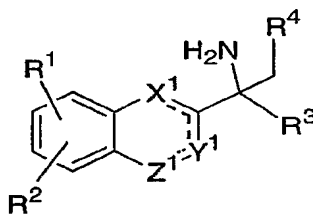
receptors could allow for an S1P mimetic that initiates platelet aggregation without affecting cell morphology.

[0009] Sphingosine-1-phosphate is formed as a metabolite of sphingosine in its reaction with sphingosine kinase and is stored in abundance in the aggregates of platelets where high levels of sphingosine kinase exist and sphingosine lyase is lacking. S1P is released during platelet aggregation, accumulates in serum, and is also found in malignant ascites. Reversible biodegradation of S1P most likely proceeds via hydrolysis by ectophosphohydrolases, specifically the sphingosine 1- phosphate phosphohydrolases. Irreversible degradation of S1P is catalyzed by S1P lyase yielding ethanolamine phosphate and hexadecenal.

[0010] Currently, there is a need for novel, potent, and selective agents that are agonists of the S1P receptor having enhanced potency, selectivity, and oral bioavailability. In addition, there is a need in the art for identification of, as well as the synthesis and use of such compounds. The present invention satisfies these needs.

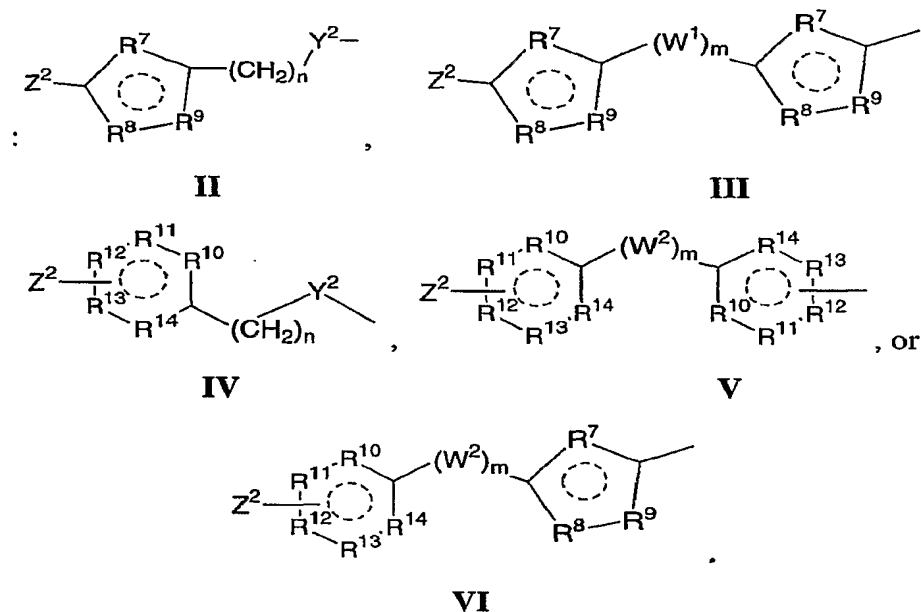
Summary

[0011] The present invention provides in one aspect compounds that have agonist activity at one or more of the S1P receptors. The compounds are sphingosine analogs that, after phosphorylation, can behave as agonists at S1P receptors. Accordingly, there is provided a compound of formula I:



I

wherein X^1 , Y^1 and Z^1 are independently O, CR^a , CR^aR^b , N, NR^c , or S; R^1 is hydrogen, halo (C₁-C₁₀)alkyl, or (C₁-C₁₀)alkoxyl; R^2 is hydrogen, halo, (C₁-C₂₀)alkyl, (C₁-C₂₀)alkoxy; (C₂-C₂₆)alkoxyalkyl; (C₂-C₂₀)alkenyl, (C₂-C₂₀)alkynyl, (C₃-C₁₂)cycloalkyl, (C₆-C₁₀)aryl, (C₇-C₃₀)arylalkyl, (C₂-C₁₀)heterocyclic, (C₄-C₁₀)heteroaryl, or R^2 can be a group having formula II, III, IV, V, or VI:



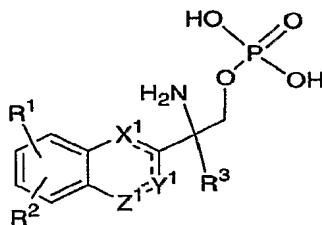
$R^7, R^8, R^9, R^{10}, R^{11}, R^{12}, R^{13}$, and R^{14} are independently O, S, C, CR^{15} , $CR^{16}R^{17}$, $C=O$, N or NR^{18} ; R^{15}, R^{16} and R^{17} are independently hydrogen, halo, (C_1-C_{10}) alkyl, (C_1-C_{10}) alkyl substituted with halo, hydroxy, (C_1-C_{10}) alkoxy, or cyano; and where R^{18} can be hydrogen or (C_1-C_{10}) alkyl and at least one of $R^{10}, R^{11}, R^{12}, R^{13}$, or R^{14} is a heteroatom (O, S or N); Z^2 is (C_1-C_6) alkyl, (C_3-C_8) cycloalkyl, substituted alkyl, (C_2-C_6) alkenyl, (C_2-C_6) alkynyl, (C_6-C_{10}) aryl, alkyl substituted aryl, (C_7-C_{16}) arylalkyl, or aryl substituted arylalkyl; wherein the alkyl groups of Z^2 are optionally substituted with 1, 2, 3, or 4 groups independently are halo, (C_1-C_{10}) alkoxy or cyano; \bigcirc indicates one or more optional double bonds; Y^2 is O, $C=O$, or CH_2 ; W^1 is a bond or $-CH_2-CH_2-CH_2-$; W^2 is a bond or $-CH_2-$ and m is 1, 2, or 3, or $(C=O)(CH_2)_{1-5}$ and m is 1. Each \equiv represents an optional double bond; n is 0, 1, 2, or 3, and q is 0, 1, 2, or 3.

[0012] R³ is hydrogen, (C₁-C₁₀)alkyl, or (C₁-C₁₀)alkoxyl; and R⁴ is hydroxyl (-OH), phosphate (-OPO₃H₂), phosphonate (-CH₂PO₃H₂), or *alpha*-substituted phosphonate; R^c is hydrogen, or (C₁-C₁₀)alkyl. R^a, R^b, and R^c are independently hydrogen, or (C₁-C₁₀)alkyl.

[0013] The alkyl groups of R¹ can be optionally substituted with 1, 2, 3, or 4 substituent groups, where the substituent groups independently are halo, (C₁-C₁₀)alkoxy or cyano. Any of the alkyl, alkenyl, alkynyl, cycloalkyl, aryl, heterocyclic, or heteroaryl groups of R² are optionally substituted with 1, 2, 3, or 4 substituent groups, where the substituent groups independently are oxo (=O), imino (=NR^d), (C₁-C₁₀)alkyl, (C₁-C₁₀)alkoxy, or C₆-aryl, or wherein one or more of the carbon atoms in the R² alkyl groups

can be independently replaced with non-peroxide oxygen, sulfur or NR^c ; the alkyl groups of R^3 are optionally substituted with 1, or 2 hydroxy groups; and R^d is hydrogen, or $(\text{C}_1\text{-C}_{10})$ alkyl. The invention includes pharmaceutically acceptable salts or esters of the compounds of formula I.

[0014] In another aspect, the invention provides phosphate esters having formula VII.



VII

[0015] In another aspect, the invention provides pro-drugs of the compounds of formula I. In another aspect, the invention also provides compounds of formula I or pharmaceutically acceptable salts or esters thereof for use in medical therapy.

[0016] In another aspect, the present invention provides a method for inhibiting angiogenesis in a tumor, comprising contacting the cancerous cells with an effective amount of a compound of formula I, or a pharmaceutically acceptable salt thereof.

[0017] In another aspect, the invention provides a method for modulating the immune system by altering lymphocyte trafficking for treatment of autoimmune diseases or prolongation of allograft transplant survival, said method comprising administering an effective amount of at least one compound of formula I to a subject in need thereof.

[0018] In another aspect, the invention provides a method for preventing, inhibiting or treating neuropathic pain, wherein the method comprises administering an effective amount of at least one compound of formula I or a compound of formula I and a pharmaceutically-acceptable carrier is administered to a subject in need thereof. Pain can be nociceptive or neuropathic in nature. Neuropathic pain is characterized by its chronic nature, an absence of an obvious, direct cause (*e.g.*, tissue damage), hyperalgesia or allodynia. Hyperalgesia is an exaggerated response to a painful stimulus. Allodynia is the perception of normal stimuli as painful (examples include the touch of clothing, warm or cool air, etc.). Neuropathic pain can be a sequel to nerve damage in an extremity such as an arm, or more often a leg. Precipitating events can include trauma, *e.g.*, motor vehicle accidents or amputations (*e.g.*, phantom limb pain). Neuropathic pain can occur due to an adverse effect of drug therapies, *e.g.*, vincristine or paclitaxel (TAXOL™) or can occur as

a component of disease pathologies, such as diabetes type 1 or type2, shingles, HIV-1 infections, etc. Typically, neuropathic pain is not responsive to opiates or non-steroidal anti-inflammatory drugs such as aspirin.

[0019] In another aspect, the invention provides a method for repairing vascular injury following catheterization, comprising contacting the lumen of the affected vessel with an effective amount of the compound of formula I. In another aspect, the invention includes coating indwelling stents with a compound of formula I.

[0020] In another aspect, the present invention provides compositions and methods for the use of S1P analogs to prevent and inhibit vascular restenosis following vascular injury. For example, the injury can be due to balloon angioplasty. In another aspect, the present invention includes a method for treating subjects to prevent vascular restenosis.

[0021] In another aspect, the present invention provides compositions and methods for the use of sphingosine analogs (including S1P pro-drugs) to prevent asthma attacks. In one aspect, the asthma could be due to over production of cysteinyl leukotrienes. In another aspect, the present invention includes a method for treating subjects to treat asthma.

[0022] In another aspect, the present invention provides compositions and methods for the use of sphingosine analogs of formula I (including S1P pro-drugs) to treat obesity.

[0023] In another aspect, the present invention provides compositions and methods for the use of sphingosine analogs (including S1P pro-drugs) to normalize blood lipid composition. In one aspect, blood low density lipoprotein (LDL or 'bad cholesterol') levels could be lowered. In another aspect, blood triglyceride levels could be lowered.

[0024] In another aspect, the present invention provides compositions and methods for the use of S1P analogs and S1P pro-drugs for the prevention and treatment of arteriosclerosis.

[0025] In another aspect, the present invention provides compositions and methods for the use of S1P analogs and S1P pro-drugs for the treatment of neoplastic disease. In one aspect, this treatment is effected by application of S1P receptor antagonists that are efficacious by virtue of their anti-angiogenic properties. In another aspect, the treatment is effected by administration of sphingosine analogs of formula I that inhibit the multiple substrate lipid kinase.

[0026] In another aspect, the present invention provides compositions and methods for the use of S1P analogs and S1P pro-drugs for the treatment of neurodegenerative diseases. In one aspect, the treatment is for senile dementia of the Alzheimers type.

[0027] In another aspect, the invention provides a compound of formula I, or a pharmaceutically acceptable salt thereof for use in medical treatment (for example, treatment of neoplastic disease, treatment of neuropathic pain, treatment of autoimmune disease, prolongation of allograft survival).

[0028] In another aspect, the invention provides a method for the use of a compound of formula I or a pharmaceutically acceptable salt thereof to prepare a medicament for inhibiting tumor growth, metastasis or tumor angiogenesis in a mammalian species (for example, a human).

[0029] In another aspect, the invention provides for the use of a compound of formula I or a pharmaceutically acceptable salt thereof to prepare a medicament for treating an autoimmune disease or prolonging allograft survival in a mammalian species (for example, a human).

[0030] In another aspect, the invention provides for the use of a compound of formula I or a pharmaceutically acceptable salt thereof to prepare a medicament for treating neuropathic pain in a mammalian species (for example, a human).

[0031] In another aspect, the invention provides a method for assessing a compound of formula I (*e.g.*, S1P receptor pro-drugs) as a substrate for sphingosine kinase types 1 or 2, *in vitro* and *in vivo*. In another aspect, the invention includes a method of assessing a compound of formula I for binding to designated receptor sites comprising *in vivo* or *in vitro*, with an amount of a compound of formula I effective to bind said receptors. Tissue comprising ligand bound designated S1P receptor sites can be used to measure the selectivity of test compounds for specific receptor subtypes, or can be used as a tool to identify potential therapeutic agents for the treatment of diseases, by contacting said agents with said ligand-receptor complexes, and measuring the extent of displacement of the ligand or binding of the agent.

[0032] In another aspect, the invention provides novel intermediates and processes disclosed herein that are useful for preparing compounds of formula I, including the generic and specific intermediates as well as the synthetic processes described herein.

[0033] In another aspect, the present invention provides synthetic schemes and methods of use of compounds having formula I and analogs or derivatives thereof. In

another aspect, the invention provides synthetic and modification schemes for preparing analogs and derivatives of the compounds of formula I, as well as compositions and methods for the use of such analogs and derivatives.

[0034] The above summary of the present invention is not intended to describe each disclosed embodiment or every implementation of the present invention. The description that follows more particularly exemplifies illustrative embodiments. In several places throughout the application, guidance is provided through lists of examples, which examples can be used in various combinations. In each instance, the recited list serves only as a representative group and should not be interpreted as an exclusive list.

[0035] The details of one or more embodiments of the invention are set forth in the accompanying description below. Other features, objects, and advantages of the invention will be apparent from the description and drawings, and from the claims.

Brief Description of the Drawings

[0036] Fig. 1 is an illustration of three S1P agonists FTY720, AA151 compound XXIX and additional compounds of formula I.

[0037] Figs. 2, 3 and 4 are schemes illustrating syntheses of compounds of formula I.

[0038] Figs. 5A, 5B and 6 illustrate the results of sphingosine kinase type 2 (SPHK2) activity for S1P agonist compounds FTY720, AA151, VIII and XVIII.

[0039] Figs. 7 and 8 graphically illustrate the results of a broken cell GTP γ ³⁵S binding assay for the human S1P₁ receptor, testing S1P and compounds XXVI, XXVII and XXVIII.

[0040] Fig. 9 is graphically illustrates total lymphocyte counts (k/ μ l) 24 hours (left bar of each group) and 48 hours (right bar of each group) after an oral dose of the test compounds are administered to mice.

[0041] Fig. 10 is graphically illustrates total lymphocyte counts (k/ μ l) 18 hours after an oral dose of the test compounds are administered to mice.

[0042] Figure 11 is a graphical representation of the results of an assay illustrating that compound VIII has no effect on the heart rate of mice. In the assay the test compound was administered via IV Dosing and the vehicle was 2% cyclodextrin.

Detailed Description

[0043] The following abbreviations are used herein: S1P, sphingosine-1-phosphate; S1P₁₋₅ S1P receptor types; GPCR, G-protein coupled receptor; SAR, structure-activity relationship; EDG, endothelial cell differentiation gene; EAE, experimental autoimmune encephalomyelitis; NOD non-obese diabetic; TNF α , tumor necrosis factor *alpha*; HDL, high density lipoprotein; and RT-PCR, reverse transcriptase polymerase chain reaction.

[0044] In describing and claiming the invention, unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Although any materials and methods similar or equivalent to those described herein can be used in the practice or testing of the present invention, the preferred materials and methods are described herein. Each of the following terms has meaning associated with it in this section. Specific and preferred values listed below for radicals, substituents, and ranges are for illustrations only; they do not exclude other defined values or other values within defined ranges for the radicals and substituents.

[0045] The terms “a,” “an,” “the,” “at least one,” and “one or more” are used interchangeably. Thus, for example, a composition that comprises “an” element means one element or more than one element.

[0046] The term “receptor agonists” are compounds that mimic the action of S1P at one or more of its receptors but may have differing potency or efficacy.

[0047] The term “receptor antagonists” are compounds that 1) lack intrinsic agonist activity and 2) block agonist (*e.g.*, S1P) activation of the S1P receptor(s), often in a manner that is both fully surmountable and reversible (‘competitive antagonist’).

[0048] The term “affected cell” refers to a cell of a subject afflicted with a disease or disorder, which affected cell has an altered phenotype relative to a subject not afflicted with a disease or disorder.

[0049] Cells or tissue are “affected” by a disease or disorder if the cells or tissue have an altered phenotype relative to the same cells or tissue in a subject not afflicted with a disease or disorder.

[0050] A disease or disorder is “alleviated” if the severity of a symptom of the disease or disorder, the frequency with which such a symptom is experienced by a patient, or both, is reduced.

[0051] An “analog” of a chemical compound is a compound that, by way of example, resembles another in structure but is not necessarily an isomer (*e.g.*, 5-fluorouracil is an analog of thymine).

[0052] The terms “cell,” “cell line,” and “cell culture” may be used interchangeably.

[0053] A “control” cell, tissue, sample, or subject is a cell, tissue, sample, or subject of the same type as a test cell, tissue, sample, or subject. The control may, for example, be examined at precisely or nearly the same time the test cell, tissue, sample, or subject is examined. The control may also, for example, be examined at a time distant from the time at which the test cell, tissue, sample, or subject is examined, and the results of the examination of the control may be recorded so that the recorded results may be compared with results obtained by examination of a test cell, tissue, sample, or subject. The control may also be obtained from another source or similar source other than the test group or a test subject, where the test sample is obtained from a subject suspected of having a disease or disorder for which the test is being performed.

[0054] A “test” cell, tissue, sample, or subject is one being examined or treated.

[0055] A “pathoindicative” cell, tissue, or sample is one which, when present, is an indication that the animal in which the cell, tissue, or sample is located (or from which the tissue was obtained) is afflicted with a disease or disorder. By way of example, the presence of one or more breast cells in a lung tissue of an animal is an indication that the animal is afflicted with metastatic breast cancer.

[0056] A tissue “normally comprises” a cell if one or more of the cell are present in the tissue in an animal not afflicted with a disease or disorder.

[0057] The use of the word “detect” and its grammatical variants is meant to refer to measurement of the species without quantification, whereas use of the word “determine” or “measure” with their grammatical variants are meant to refer to measurement of the species with quantification. The terms “detect” and “identify” are used interchangeably herein.

[0058] A “detectable marker” or a “reporter molecule” is an atom or a molecule that permits the specific detection of a compound comprising the marker in the presence of similar compounds without a marker. Detectable markers or reporter molecules include, *e.g.*, radioactive isotopes, antigenic determinants, enzymes, nucleic acids available for hybridization, chromophores, fluorophores, chemiluminescent molecules,

electrochemically detectable molecules, and molecules that provide for altered fluorescence-polarization or altered light-scattering.

[0059] A “disease” is a state of health of an animal wherein the animal cannot maintain homeostasis, and wherein if the disease is not ameliorated then the animal's health continues to deteriorate.

[0060] A “disorder” in an animal is a state of health in which the animal is able to maintain homeostasis, but in which the animal's state of health is less favorable than it would be in the absence of the disorder. Left untreated, a disorder does not necessarily cause a further decrease in the animal's state of health.

[0061] An “effective amount” means an amount sufficient to produce a selected effect. For example, an effective amount of an S1P receptor antagonist is an amount that decreases the cell signaling activity of the S1P receptor.

[0062] A “functional” molecule is a molecule in a form in which it exhibits a property by which it is characterized. By way of example, a functional enzyme is one which exhibits the characteristic catalytic activity by which the enzyme is characterized.

[0063] The term “inhibit” refers to the ability of a disclosed compound to reduce or impede a described function. Preferably, inhibition is by at least 10%, more preferably by at least 25%, even more preferably by at least 50%, and most preferably, the function is inhibited by at least 75%.

[0064] “Instructional material” includes a publication, a recording, a diagram, or any other medium of expression which can be used to communicate the usefulness of the disclosed compounds in the kit for effecting alleviation of the various diseases or disorders recited herein. Optionally, or alternately, the instructional material may describe one or more methods of alleviating the diseases or disorders in a cell or a tissue of a mammal. The instructional material of the kit may, for example, be affixed to a container which contains a disclosed compound or be shipped together with a container which contains the identified compound. Alternatively, the instructional material may be shipped separately from the container with the intention that the instructional material and the compound be used cooperatively by the recipient.

[0065] The term “parenteral” means not through the alimentary canal but by some other route such as subcutaneous, intramuscular, intraspinal, or intravenous.

[0066] The term “pharmaceutically acceptable carrier” includes any of the standard pharmaceutical carriers, such as a phosphate buffered saline solution, water and emulsions such as an oil/water or water/oil emulsion, and various types of wetting agents. The term also encompasses any of the agents approved by a regulatory agency of the U.S. Federal government or listed in the U.S. Pharmacopeia for use in animals, including humans.

[0067] The term “purified” and similar terms relate to the isolation of a molecule or compound in a form that is substantially free (at least 75% free, preferably 90% free, and most preferably at least 95% free) from other components normally associated with the molecule or compound in a native environment. The term “purified” does not necessarily indicate that complete purity of the particular molecules achieved during the process. A “very pure” compound refers to a compound that is greater than 90% pure. A “highly purified” compound refers to a compound that is greater than 95% pure.

[0068] A “sample” refers preferably to a biological sample from a subject, including, but not limited to, normal tissue samples, diseased tissue samples, biopsies, blood, saliva, feces, semen, tears, and urine. A sample can also be any other source of material obtained from a subject, which contains cells, tissues, or fluid of interest. A sample can also be obtained from cell or tissue culture.

[0069] The term “standard,” refers to something used for comparison. For example, a standard can be a known standard agent or compound which is administered or added to a control sample and used for comparing results when measuring said compound in a test sample. Standard can also refer to an “internal standard,” such as an agent or compound which is added at known amounts to a sample and is useful in determining such things as purification or recovery rates when a sample is processed or subjected to purification or extraction procedures before a marker of interest is measured.

[0070] A “subject” of analysis, diagnosis, or treatment is an animal. Such animals include mammals, preferably a human.

[0071] A “therapeutic” treatment is a treatment administered to a subject who exhibits signs of pathology for the purpose of diminishing or eliminating those signs.

[0072] A “therapeutically effective amount” of a compound is that amount of compound which is sufficient to provide a beneficial effect to the subject to which the compound is administered.

[0073] The term “treating” includes prophylaxis of the specific disorder or condition, or alleviation of the symptoms associated with a specific disorder or condition or preventing or eliminating said symptoms.

[0074] The disclosed compounds are generally named according to the IUPAC or CAS nomenclature system. Abbreviations which are well known to one of ordinary skill in the art may be used (*e.g.*, “Ph” for phenyl, “Me” for methyl, “Et” for ethyl, “h” for hour or hours, “rt” for room temperature, and “*rac*” for racemic mixture).

[0075] The values listed below for radicals, substituents, and ranges, are for illustration only; they do not exclude other defined values or other values within defined ranges for the radicals and substituents. The disclosed compounds include compounds of formula I having any combination of the values, specific values, more specific values, and preferred values described herein.

[0076] The term “halogen” or “halo” includes bromo, chloro, fluoro, and iodo. The term “haloalkyl”, refers to an alkyl radical bearing at least one halogen substituent, non-limiting examples include, but are not limited to, chloromethyl, fluoroethyl or trifluoromethyl and the like. The term “C₁-C₂₀ alkyl” refers to a branched or linear alkyl group having from one to nine carbons. Non-limiting examples include, but are not limited to, methyl, ethyl, *n*-propyl, iso-propyl, butyl, iso-butyl, sec-butyl, tert-butyl, pentyl, hexyl, heptyl, octyl and the like. The term “C₂-C₂₀ alkenyl”, refers to an olefinically unsaturated branched or linear group having from two to nine carbon atoms and at least one double bond. Typically, C₂-C₂₀ alkenyl groups include, but are not limited to, 1-propenyl, 2-propenyl, 1,3-butadienyl, 1-butenyl, hexenyl, pentenyl, hexenyl, heptenyl, octenyl and the like. The term (C₂-C₂₀)alkynyl can be ethynyl, 1-propynyl, 2-propynyl, 1-butylnyl, 2-butylnyl, 3-butylnyl, 1-pentylnyl, 2-pentylnyl, 3-pentylnyl, 4-pentylnyl, 1-hexynyl, 2-hexynyl, 3-hexynyl, 4-hexynyl, or 5-hexynyl, and the like. The term “(C₁-C₁₀)alkoxy or alkoxyl” refers to an alkyl group attached through an oxygen atom. Examples of (C₁-C₁₀)alkoxy can be methoxy, ethoxy, propoxy, isopropoxy, butoxy, isobutoxy, sec-butoxy, pentoxy, 3-pentoxy, or hexyloxy and the like. The term “C₃-C₁₂ cycloalkyl”, can be cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, cyclooctyl and the like.

[0077] The term “optionally substituted” refers to zero, one, two, three or four substituents, wherein the substituents are each independently selected. Each of the independently selected substituents may be the same or different than other substituents.

[0078] The term “(C₆-C₁₀)aryl” refers to a mono or bicyclic carbocyclic ring system having one or two aromatic rings including, but not limited to, phenyl, benzyl, naphthyl, tetrahydronaphthyl, indanyl, indenyl, and the like.

[0079] The term “aryl(C₁-C₂₀)alkyl” or “aralkyl” refers to an alkyl group substituted with a mono or bicyclic carbocyclic ring system having one or two aromatic rings including, a group such as phenyl, naphthyl, tetrahydronaphthyl, indanyl, indenyl, and the like. Non-limiting examples of arylalkyl include benzyl, phenylethyl, and the like.

[0080] The term “optionally substituted aryl” includes aryl compounds having zero, one, two, three or four substituents, and a substituted aryl includes aryl compounds having one, two, three or four substituents, wherein the substituents include groups such as, for example, alkyl, halo, or amino substituents.

[0081] The “(C₂-C₁₀)heterocyclic group” refers to an optionally substituted mono- or bicyclic carbocyclic ring system containing one, two, or three heteroatoms (optionally in each ring) wherein the heteroatoms are oxygen, sulfur, and nitrogen.

[0082] The term “(C₄-C₁₀)heteroaryl” refers to an optionally substituted mono- or bicyclic carbocyclic ring system containing one, two, or three heteroatoms (optionally in each ring) wherein the heteroatoms are oxygen, sulfur, and nitrogen. Non-limiting examples of heteroaryl groups include furyl, thienyl, pyridyl, and the like.

[0083] The term “bicyclic” represents either an unsaturated or saturated stable bridged or fused bicyclic carbon ring. The bicyclic ring may be attached at any carbon atom which affords a stable structure. Typically a bicyclic ring system can have from about 7- to about 12 atoms in the ring system. The term includes, but is not limited to, naphthyl, dicyclohexyl, dicyclohexenyl, and the like.

[0084] The term “phosphate analog” and “phosphonate analog” comprise analogs of phosphate and phosphonate wherein the phosphorous atom is in the +5 oxidation state and one or more of the oxygen atoms is replaced with a non-oxygen moiety, including for example, the phosphate analogs phosphorothioate, phosphorodithioate, phosphoroselenoate, phosphorodiselenoate, phosphoroanilothioate, phosphoranilidate, phosphoramidate, boronophosphates, and the like, including associated counterions, *e.g.*, H, NH₄, Na, K, and the like if such counterions are present.

[0085] A “derivative” of a compound refers to a chemical compound that may be produced from another compound of similar structure in one or more steps, such as replacement of hydrogen by an alkyl, acyl, or amino group.

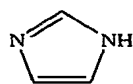
[0086] The term “pharmaceutically acceptable carrier” includes any of the standard pharmaceutical carriers, such as a phosphate buffered saline solution, hydroxypropyl beta-cyclodextrins (HO-propyl beta cyclodextrins), water, emulsions such as an oil/water or water/oil emulsion, and various types of wetting agents. The term also encompasses any of the agents approved by a regulatory agency of the U.S. Federal government or listed in the U.S. Pharmacopeia for use in animals, including humans.

[0087] The term “pharmaceutically-acceptable salt” refers to salts which retain the biological effectiveness and properties of the disclosed compounds and which are not biologically or otherwise undesirable. In many cases, the disclosed compounds are capable of forming acid or base salts by virtue of the presence of amino or carboxyl groups or groups similar thereto.

[0088] An “effective amount” means an amount sufficient to produce a selected effect. For example, an effective amount of an S1P receptor agonist is an amount that decreases the cell signaling activity of the S1P receptor.

[0089] The disclosed compounds can contain one or more asymmetric centers in the molecule. In accordance with the present disclosure any structure that does not designate the stereochemistry is to be understood as embracing all the various optical isomers, as well as racemic mixtures thereof.

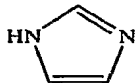
[0090] The disclosed compounds may exist in tautomeric forms and the invention includes both mixtures and separate individual tautomers. For example, the following structure:



is understood to represent a mixture of the structures:



as well as



[0091] The terms 16:0, 18:0, 18:1, 20:4 or 22:6 hydrocarbon refers to a branched or straight alkyl or alkenyl group, wherein the first integer represents the total number of carbons in the group and the second integer represent the number of double bonds in the group.

[0092] An “S1P modulating agent” refers a compound or composition that is capable of inducing a detectable change in S1P receptor activity *in vivo* or *in vitro* (e.g., at least 10% increase or decrease in S1P activity as measured by a given assay such as the

bioassay described in the examples and known in the art. "S1P receptor," refers to all of the S1P receptor subtypes (for example, the S1P receptors S1P₁, S1P₂, S1P₃, S1P₄, and S1P₅), unless the specific subtype is indicated.

[0093] It will be appreciated by those skilled in the art that the disclosed compounds having chiral centers may exist in and be isolated in optically active and racemic forms. It is to be understood that the disclosed compounds encompass any racemic, optically active or stereoisomeric form, or mixtures thereof, of the compound, which possess the useful properties described herein, such as the *S,R*; *S,S*; *R,R*; or *R,S* diastereomers. It is well known in the art how to prepare such optically active forms (for example, by resolution of the racemic form by recrystallization techniques, by synthesis from optically-active starting materials, by chiral synthesis, or by chromatographic separation using a chiral stationary phase) and how to determine S1P agonist activity using the standard tests described herein, or using other similar tests which are well known in the art. In addition, some compounds may exhibit polymorphism.

[0094] Potential uses of an S1P receptor agonist pro-drugs (S1P₁ receptor type selective agonists preferred) include, but are not limited to, altering lymphocyte trafficking as a method of treatment for autoimmune pathologies such as uveitis, type I diabetes, rheumatoid arthritis, inflammatory bowel diseases, and, most particularly, multiple sclerosis. "Treatment" of multiple sclerosis includes the various forms of the disease including relapsing-remitting, chronic progressive, etc., and the S1P receptor agonists can be used alone or in conjunction with other agents to relieve signs and symptoms of the disease as well as prophylactically.

[0095] In addition, the disclosed compounds can be used for altering lymphocyte trafficking as a method for prolonging allograft survival, for example solid organ transplants, treatment of graft vs. host disease, bone marrow transplantation, and the like.

[0096] In addition, the disclosed compounds can be used to inhibit autotaxin. Autotaxin, a plasma phosphodiesterase, has been demonstrated to undergo end product inhibition. Autotaxin hydrolyzes several substrates to yield lysophosphatidic acid and sphingosine 1-phosphate, and has been implicated in cancer progression and angiogenesis. Therefore, S1P receptor agonist pro-drugs of the disclosed compounds can be used to inhibit autotaxin. This activity may be combined with agonism at S1P receptors or may be independent of such activity.

[0097] In addition, disclosed compounds can be useful for inhibition of S1P lyase. S1P lyase is an intracellular enzyme that irreversibly degrades S1P. Inhibition of S1P lyase disrupts lymphocyte trafficking with concomitant lymphopenia. Accordingly, S1P lyase inhibitors can be useful in modulating immune system function. Therefore, the disclosed compounds can be used to inhibit S1P lyase. This inhibition could be in concert with S1P receptor activity, or be independent of activity at any S1P receptor.

[0098] In addition, disclosed compounds can be useful as antagonists of the cannabinoid CB₁ receptor. CB₁ antagonism is associated with a decrease in body weight and an improvement in blood lipid profiles. The CB₁ antagonism could be in concert with S1P receptor activity, or be independent of activity at any S1P receptor.

[0099] In addition, disclosed compounds can be useful for inhibition of group IVA cytosolic PLA₂ (cPLA₂). cPLA₂ catalyzes the release of eicosanoic acids (*e.g.*, arachidonic acid). The eicosanoic acids are transformed to pro-inflammatory eicosanoids such as prostaglandins and leukotrienes. Thus, disclosed compounds may be useful as anti-inflammatory agents. This inhibition could be in concert with S1P receptor activity, or be independent of activity at any S1P receptor.

[00100] In addition, disclosed compounds may be useful for inhibition of the multiple substrate lipid kinase (MuLK). MuLK is highly expressed in many human tumor cells and thus its inhibition might slow the growth or spread of tumors.

[00101] "Treatment" of multiple sclerosis includes the various forms of the disease including relapsing-remitting, chronic progressive, etc., and the S1P receptor agonists can be used alone or in conjunction with other agents to relieve signs and symptoms of the disease as well as prophylactically.

[00102] The present invention is also includes pharmaceutical compositions comprising the compounds of formula I. More particularly, such compounds can be formulated as pharmaceutical compositions using standard pharmaceutically acceptable carriers, fillers, solubilizing agents and stabilizers known to those skilled in the art. For example, a pharmaceutical composition comprising a compound of formula I, or analog, derivative, or modification thereof, as described herein, is used to administer the appropriate compound to a subject.

[00103] The compounds of formula I are useful for treating a disease or disorder including administering to a subject in need thereof of a therapeutically acceptable amount

of a compound of formula I, or a pharmaceutical composition comprising a therapeutically effective amount of a compound of formula I, and a pharmaceutically-acceptable carrier.

[00104] The disclosed compounds and method are directed to sphingosine 1-phosphate (S1P) analogs that have activity as receptor agonists or antagonists at one or more S1P receptors, specifically the S1P₁, S1P₄ and S1P₅ receptor types. The disclosed compounds and method include both compounds that have a phosphate moiety as well as compounds with hydrolysis-resistant phosphate surrogates such as phosphonates, *alpha*-substituted phosphonates particularly where the *alpha* substitution is a halogen and phosphothionates.

[00105] The values listed below for radicals, substituents, and ranges, are for illustration only; they do not exclude other defined values or other values within defined ranges for the radicals and substituents.

[00106] X¹, Y¹ and Z¹ are independently O, CH, CH₂, CHCF₃, N, NH, or S.

[00107] Another value for X^1 , Y^1 and Z^1 is CH_2 .

[00108] R¹ can be hydrogen, fluorine, chlorine, bromine, trifluoromethyl, methoxy, (C₁-C₆)alkyl, (C₁-C₆)haloalkyl, or (C₁-C₆)alkyl substituted with, alkoxy or cyano.

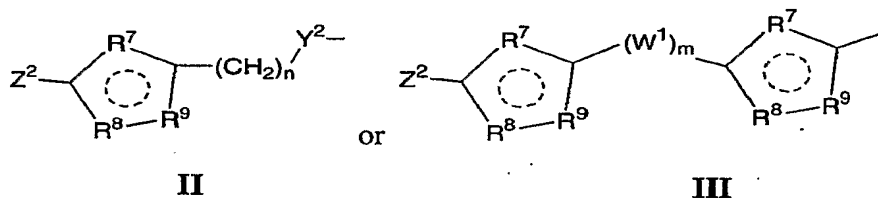
[00109] Additional values for R¹ are hydrogen, trifluoromethyl, or -CH₂CF₃.

[00110] More additional values for R¹ are alkyl-substituted aryl, aryl-substituted alkyl, or aryl-substituted arylalkyl.

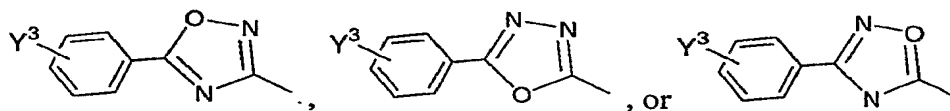
[00111] More additional values for R¹ are benzyl, phenylethyl, or methyl benzyl.

[00112] Compounds having formula I can have an R² group that includes a chain having the structure -CH₂-CH₂-O-CH₂-CH₂-O-.

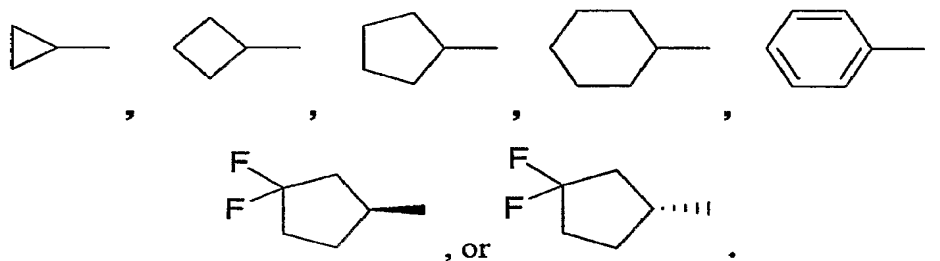
[00113] Values for R^2 include



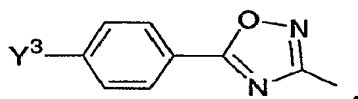
[00114] Additional values for R^2 having formula II are



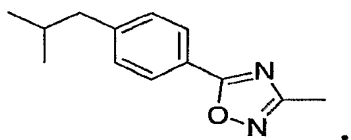
where Y^3 is $(CH_3)_3C-$, $CH_3CH_2(CH_3)_2C-$, $CH_3CH_2CH_2-$, $CH_3(CH_2)_2CH_2-$, $CH_3(CH_2)_4CH_2-$, $(CH_3)_2CHCH_2-$, $(CH_3)_3CCH_2-$, CH_3CH_2O- , $(CH_3)_2CHO-$, or $CF_3CH_2CH_2-$ or a group having the formula:



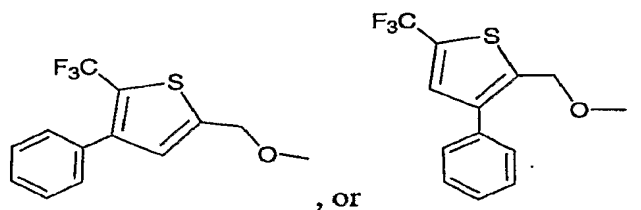
[00115] An additional value for R^2 having formula II (*para* substituted 3,5-diphenyl-(1,2,4)-oxadiazole) is;



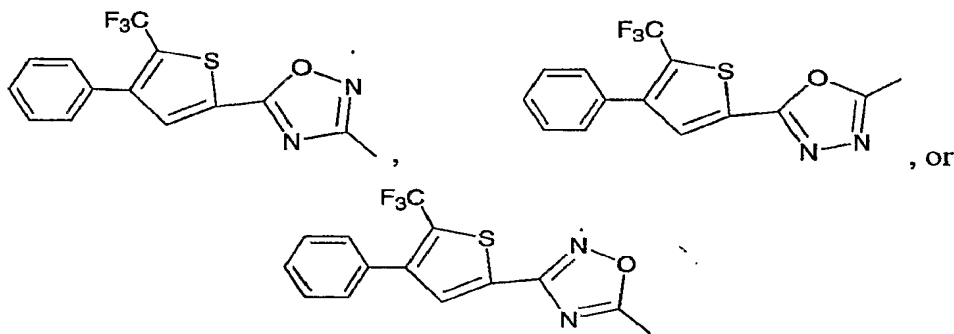
[00116] Another value for R^2 having formula II is;



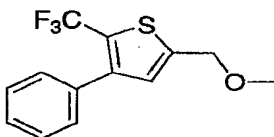
[00117] Another value for R^2 having formula II is;



[00118] Additional values for R^2 having formula III are;



[00119] Another value for R^2 having formula III is;



[00120] Additional values for R^2 include (C_1-C_{20}) alkyl, (C_1-C_{20}) alkoxy, or (C_2-C_{26}) alkoxyalkyl.

[00121] More additional values for R^2 include (C_1-C_{10}) alkyl, (C_2-C_{10}) alkenyl and (C_2-C_{14}) alkynyl or (C_1-C_{10}) alkoxy optionally substituted with carbonyl ($C=O$) or oxime ($C=NR^d$) groups.

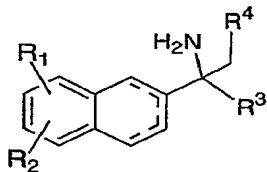
[00122] Additional values for R^2 include methyl, ethyl, propyl, butyl, pentyl, hexyl, heptyl, octyl, trifluoromethyl, trifluoroethyl, trifluoromethoxy, trifluoroethoxy, methoxy, ethoxy, propoxy, butoxy, pentoxy, heptoxy, or octoxy.

[00123] Another value for R^3 is methyl, hydroxymethyl, ethyl, hydroxyethyl, propyl, hydroxypropyl, or isopropyl.

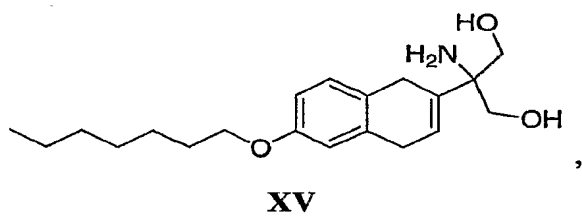
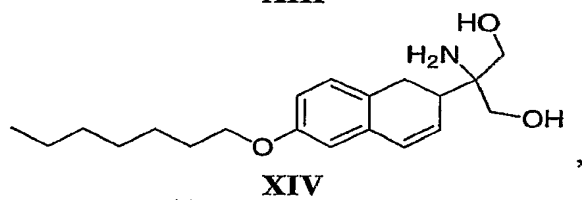
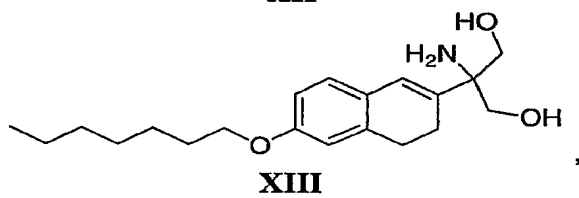
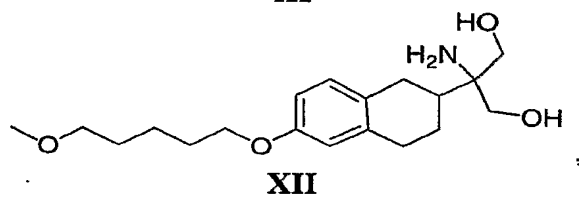
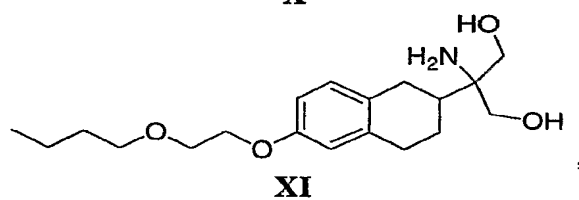
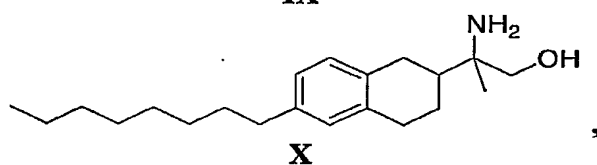
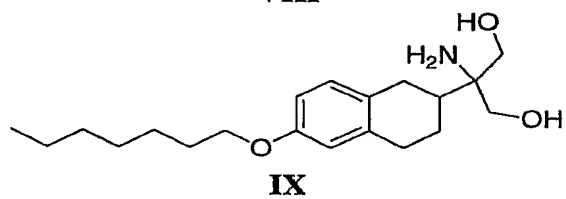
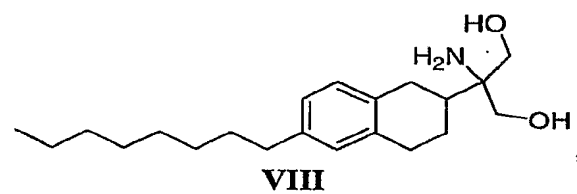
[00124] Another value for R^3 is methyl, hydroxymethyl, ethyl, or hydroxyethyl.

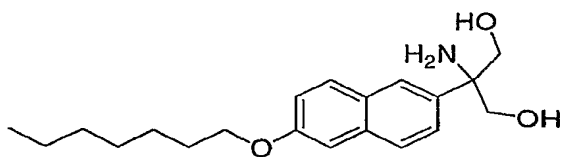
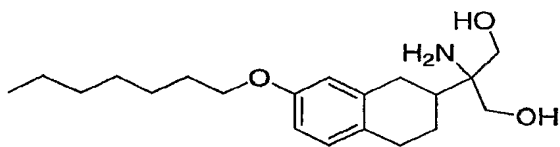
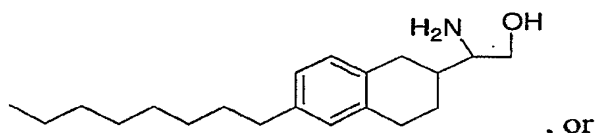
[00125] A value for R^4 is hydroxy, or phosphate ($-OPO_3H_2$).

[00126] A specific compound has the formula

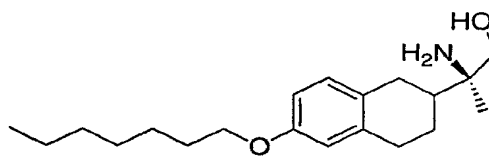


[00127] Additional compounds have formulas



**XVI****XVII**

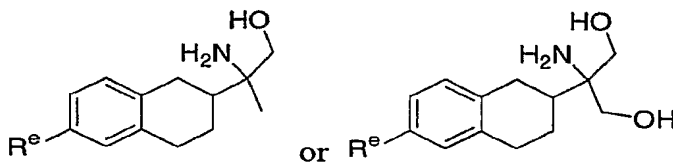
, or

XVIII**XIX**

[00128] Additional compounds having formula I include compounds above or in Fig. 1 where one of more of the hydrogen atoms from a hydroxy group is replaced with a phosphate group --OP(=O)(OH)_2 .

[00129] Additional compounds of formula I are illustrated in table 1, below.

Table 1



Compound	R ^e
XX	
XXI	
XXII	
XXIII	
XXIV	
XXV	

[00130] In another aspect, the invention provides S1P receptor pro-drug compounds having the general structure of formula I, is provided by a compound with a mono-substituted tetralin ring system that has the formula VIII. In some embodiments of formula I, the structure (*e.g.*, IX and X) has only a single chiral center and that the amino carbon is pro-chiral, *e.g.*, will become chiral following enzyme-catalyzed phosphorylation.

[00131] Without wishing to be bound by any particular theory, it is expected that the compounds described herein are pro-drugs, *e.g.*, are activated by phosphorylation of the primary alcohol to form the mono-phosphorylated analog. Additionally, the active drugs are expected to be agonists at the S1P type 1 receptor.

[00132] In cases where compounds of formula I are sufficiently basic or acidic to form stable nontoxic acid or base salts, preparation and administration of the compounds as pharmaceutically acceptable salts may be appropriate. Examples of pharmaceutically acceptable salts are organic acid addition salts formed with acids which form a physiological acceptable anion, for example, tosylate, methanesulfonate, acetate, citrate, malonate, tartarate, succinate, benzoate, ascorbate, α -ketoglutarate, and α -glycerophosphate. Inorganic salts may also be formed, including hydrochloride, sulfate, nitrate, bicarbonate, and carbonate salts.

[00133] Pharmaceutically acceptable salts may be obtained using standard procedures well known in the art, for example by reacting a sufficiently basic compound such as an amine with a suitable acid affording a physiologically acceptable anion. Alkali metal (for example, sodium, potassium or lithium) or alkaline earth metal (for example calcium) salts of carboxylic acids can also be made.

[00134] Pharmaceutically-acceptable base addition salts can be prepared from inorganic and organic bases. Salts from inorganic bases, include but are not limited to, sodium, potassium, lithium, ammonium, calcium and magnesium salts. Salts derived from organic bases include, but are not limited to, salts of primary, secondary and tertiary amines, such as alkyl amines, dialkyl amines, trialkyl amines, substituted alkyl amines, di(substituted alkyl) amines, tri(substituted alkyl) amines, alkenyl amines, dialkenyl amines, trialkenyl amines, substituted alkenyl amines, di(substituted alkenyl) amines, tri(substituted alkenyl) amines, cycloalkyl amines, di(cycloalkyl) amines, tri(cycloalkyl) amines, substituted cycloalkyl amines, disubstituted cycloalkyl amine, trisubstituted cycloalkyl amines, cycloalkenyl amines, di(cycloalkenyl) amines, tri(cycloalkenyl) amines, substituted cycloalkenyl amines, disubstituted cycloalkenyl amine, trisubstituted cycloalkenyl amines, aryl amines, diaryl amines, triaryl amines, heteroaryl amines, diheteroaryl amines, triheteroaryl amines, heterocyclic amines, diheterocyclic amines, triheterocyclic amines, mixed di- and tri-amines where at least two of the substituents on the amine are different and are alkyl, substituted alkyl, alkenyl, substituted alkenyl, cycloalkyl, substituted cycloalkyl, cycloalkenyl, substituted cycloalkenyl, aryl, heteroaryl, or heterocyclic and the

like. Also included are amines where the two or three substituents, together with the amino nitrogen, form a heterocyclic or heteroaryl group. Non-limiting examples of amines include, isopropylamine, trimethyl amine, diethyl amine, tri(iso-propyl) amine, tri(n-propyl) amine, ethanolamine, 2-dimethylaminoethanol, tromethamine, lysine, arginine, histidine, caffeine, procaine, hydrabamine, choline, betaine, ethylenediamine, glucosamine, N-alkylglucamines, theobromine, purines, piperazine, piperidine, morpholine, N-ethylpiperidine, and the like. It should also be understood that other carboxylic acid derivatives would be useful, for example, carboxylic acid amides, including carboxamides, lower alkyl carboxamides, dialkyl carboxamides, and the like.

[00135] The compounds of formula I can be formulated as pharmaceutical compositions and administered to a mammalian host, such as a human patient in a variety of forms adapted to the chosen route of administration, *e.g.*, orally or parenterally, by intravenous, intramuscular, topical or subcutaneous routes.

[00136] Thus, the present compounds may be systemically administered, *e.g.*, orally, in combination with a pharmaceutically acceptable vehicle such as an inert diluent or an assimilable edible carrier. They may be enclosed in hard or soft shell gelatin capsules, may be compressed into tablets, or may be incorporated directly with the food of the patient's diet. For oral therapeutic administration, the active compound may be combined with one or more excipients and used in the form of ingestible tablets, buccal tablets, troches, capsules, elixirs, suspensions, syrups, wafers, and the like. Such compositions and preparations should contain at least about 0.1% of active compound. The percentage of the compositions and preparations may, of course, be varied and may conveniently be between about 2 to about 60% of the weight of a given unit dosage form. The amount of active compound in such therapeutically useful compositions is such that an effective dosage level will be obtained.

[00137] The tablets, troches, pills, capsules, and the like may also contain the following: binders such as gum tragacanth, acacia, corn starch or gelatin; excipients such as dicalcium phosphate; a disintegrating agent such as corn starch, potato starch, alginic acid and the like; a lubricant such as magnesium stearate; and a sweetening agent such as sucrose, fructose, lactose or aspartame or a flavoring agent such as peppermint, oil of wintergreen, or cherry flavoring may be added. When the unit dosage form is a capsule, it may contain, in addition to materials of the above type, a liquid carrier, such as a vegetable oil or a polyethylene glycol. Various other materials may be present as coatings or to

otherwise modify the physical form of the solid unit dosage form. For instance, tablets, pills, or capsules may be coated with gelatin, wax, shellac or sugar and the like. A syrup or elixir may contain the active compound, sucrose or fructose as a sweetening agent, methyl and propylparabens as preservatives, a dye and flavoring such as cherry or orange flavor. Of course, any material used in preparing any unit dosage form should be pharmaceutically acceptable and substantially non-toxic in the amounts employed. In addition, the active compound may be incorporated into sustained-release preparations and devices.

[00138] The active compound may also be administered intravenously or intraperitoneally by infusion or injection. Solutions of the active compound or its salts can be prepared in water, optionally mixed with a nontoxic surfactant. Dispersions can also be prepared in glycerol, liquid polyethylene glycols, triacetin, and mixtures thereof and in oils. Under ordinary conditions of storage and use, these preparations contain a preservative to prevent the growth of microorganisms.

[00139] Exemplary pharmaceutical dosage forms for injection or infusion can include sterile aqueous solutions or dispersions or sterile powders comprising the active ingredient which are adapted for the extemporaneous preparation of sterile injectable or infusible solutions or dispersions, optionally encapsulated in liposomes. In all cases, the ultimate dosage form should be sterile, fluid and stable under the conditions of manufacture and storage. The liquid carrier or vehicle can be a solvent or liquid dispersion medium comprising, for example, water, ethanol, a polyol (for example, glycerol, propylene glycol, liquid polyethylene glycols, and the like), vegetable oils, nontoxic glyceryl esters, and mixtures thereof. The proper fluidity can be maintained, for example, by the formation of liposomes, by the maintenance of the required particle size in the case of dispersions or by the use of surfactants. The prevention of the action of microorganisms can be brought about by various antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, sorbic acid, thimerosal, and the like. In many cases, it will be preferable to include isotonic agents, for example, sugars, buffers or sodium chloride. Prolonged absorption of the injectable compositions can be brought about by the use in the compositions of agents delaying absorption, for example, aluminum monostearate and gelatin.

[00140] Sterile injectable solutions are prepared by incorporating the active compound in the required amount in the appropriate solvent with various of the other ingredients

enumerated above, as required, followed by filter sterilization. In the case of sterile powders for the preparation of sterile injectable solutions, the preferred methods of preparation are vacuum drying and the freeze drying techniques, which yield a powder of the active ingredient plus any additional desired ingredient present in the previously sterile-filtered solutions.

[00141] For topical administration, the present compounds may be applied in pure form, *e.g.*, when they are liquids. However, it will generally be desirable to administer them to the skin as compositions or formulations, in combination with a dermatologically acceptable carrier, which may be a solid or a liquid.

[00142] Exemplary solid carriers include finely divided solids such as talc, clay, microcrystalline cellulose, silica, alumina and the like. Useful liquid carriers include water, alcohols or glycols or water-alcohol/glycol blends, in which the present compounds can be dissolved or dispersed at effective levels, optionally with the aid of non-toxic surfactants. Adjuvants such as fragrances and additional antimicrobial agents can be added to optimize the properties for a given use. The resultant liquid compositions can be applied from absorbent pads, used to impregnate bandages and other dressings, or sprayed onto the affected area using pump-type or aerosol sprayers.

[00143] Thickeners such as synthetic polymers, fatty acids, fatty acid salts and esters, fatty alcohols, modified celluloses or modified mineral materials can also be employed with liquid carriers to form spreadable pastes, gels, ointments, soaps, and the like, for application directly to the skin of the user.

[00144] Examples of useful dermatological compositions which can be used to deliver the compounds of formula I to the skin are known to the art; for example, see Jacquet *et al.* (U.S. Pat. No. 4,608,392), Geria (U.S. Pat. No. 4,992,478), Smith *et al.* (U.S. Pat. No. 4,559,157) and Wortzman (U.S. Pat. No. 4,820,508).

[00145] Useful dosages of the compounds of formula I can be determined by comparing their *in vitro* activity, and *in vivo* activity in animal models. Methods for the extrapolation of effective dosages in mice, and other animals, to humans are known to the art; for example, see U.S. Pat. No. 4,938,949.

[00146] Generally, the concentration of the compound(s) of formula I in a liquid composition, such as a lotion, will be from about 0.1 to about 25 weight percent, preferably from about 0.5-10 weight percent. The concentration in a semi-solid or solid

composition such as a gel or a powder will be about 0.1-5 wt-%, preferably about 0.5-2.5 weight percent based on the total weight of the composition.

[00147] The amount of the compound, or an active salt or derivative thereof, required for use in treatment will vary not only with the particular salt selected but also with the route of administration, the nature of the condition being treated and the age and condition of the patient and will be ultimately at the discretion of the attendant physician or clinician. In general, however, a dose will be in the range of from about 0.1 to about 10 mg/kg of body weight per day.

[00148] The compound is conveniently administered in unit dosage form; for example, containing 5 to 1000 mg, conveniently 10 to 750 mg, most conveniently, 50 to 500 mg of active ingredient per unit dosage form.

[00149] Ideally, the active ingredient should be administered to achieve peak plasma concentrations of the active compound of from about 0.5 to about 75 μM , preferably, about 1 to 50 μM , most preferably, about 2 to about 30 μM . This may be achieved, for example, by the intravenous injection of a 0.05 to 5% solution of the active ingredient, optionally in saline, or orally administered as a bolus containing about 1-100 mg of the active ingredient. Desirable blood levels may be maintained by continuous infusion to provide about 0.01-5.0 mg/kg/hr or by intermittent infusions containing about 0.4-15 mg/kg of the active ingredient(s).

[00150] The desired dose may conveniently be presented in a single dose or as divided doses administered at appropriate intervals, for example, as two, three, four, or more sub-doses per day. The sub-dose itself may be further divided, *e.g.*, into a number of discrete loosely spaced administrations; such as multiple inhalations from an insufflator or by application of a plurality of drops into the eye.

[00151] The disclosed method includes a kit comprising an inhibitor compound of formula I and instructional material which describes administering the inhibitor compound or a composition comprising the inhibitor compound to a cell or a subject. This should be construed to include other embodiments of kits that are known to those skilled in the art, such as a kit comprising a (preferably sterile) solvent for dissolving or suspending the inhibitor compound or composition prior to administering the compound or composition to a cell or a subject. Preferably, the subject is a human.

[00152] In accordance with the disclosed compounds and methods, as described above or as discussed in the Examples below, there can be employed conventional chemical,

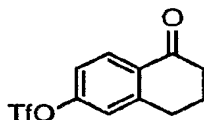
cellular, histochemical, biochemical, molecular biology, microbiology, and *in vivo* techniques which are known to those of skill in the art. Such techniques are explained fully in the literature.

[00153] Without further description, it is believed that one of ordinary skill in the art can, using the preceding description and the following illustrative examples, make and utilize the disclosed compounds.

[00154] Processes for preparing compounds of formula I or for preparing intermediates useful for preparing compounds of formula I are provided as further embodiments. Intermediates useful for preparing compounds of formula I are also provided as further embodiments. The processes are provided as further embodiments and are illustrated in the schemes herein wherein the meanings of the generic radicals are as given above unless otherwise qualified.

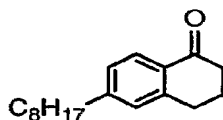
[00155] An example of the synthesis of several disclosed compounds is illustrated in Scheme 1 (Fig. 2). The reagents and conditions are as follows: a) TiCl_4 , 2,6-lutidine, CH_2Cl_2 , 0°C , 2h, 93%; b) 1-octene, 9-BBN, K_3PO_4 , KBr, H_2O , $\text{Pd}(\text{PPh}_3)_4$, 65°C , 2h, 82%; c) CuBr_2 , EtOAc, CHCl_3 , reflux 6h, 80%; d) NaH, *N*-acetamido-diethylmalonate, DMF, 0°C -rt, overnight, 75%; e) Et_3SH , TiCl_4 , CH_2Cl_2 , rt, 12h, 65%; f) LiBH_4 , rt, THF, 48h, 33%; g) LiOH, H_2O , MeOH, THF, 50°C , 5h, 75%; h) P_2O_5 , H_3PO_4 , 100°C , 2h, 37%; i) 12M HCL, MeOH, reflux, 2h; j) LiAlH_4 , THF, reflux, 12h, 21%, two steps; k) P_2O_5 , H_3PO_4 , 100°C , 2h, 50%.

[00156] The invention is now described with reference to the following Examples and Embodiments. Without further description, it is believed that one of ordinary skill in the art can, using the preceding description and the following illustrative examples, make and utilize the disclosed compounds. The following working examples therefore, are provided for the purpose of illustration only and specifically point out the preferred embodiments, and are not to be construed as limiting in any way the remainder of the disclosure. Therefore, the examples should be construed to encompass any and all variations which become evident as a result of the teaching provided herein.

ExamplesExample 1: Trifluoromethanesulfonic acid 5-oxo-5,6,7,8-tetrahydro-naphthalen-2-yl ester (2).

[00157] Trifluoromethanesulfonic anhydride (1.7 mL, 10 mmol) was added slowly over 1 hour to a solution of 6-hydroxy-1-tetralone (1.62 g, 10 mmol) and 2,6-lutidine (1.28 mL, 10 mmol) in dry dichloromethane (10 mL) cooled to 0 °C. After 1 hour the solution was diluted with dichloromethane (10 mL) and washed with 1 M hydrochloric acid (20 mL). The organic layer was re-extracted with dichloromethane (50 mL) and the combined organics washed with 1 M hydrochloric acid (10 mL). The organics were dried over magnesium sulfate and concentrated under vacuum. The residue was purified by column chromatography (Silica gel, CH₂Cl₂) to provide 2.7 g of compound **2** (93%).

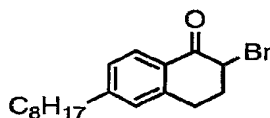
[00158] ¹H NMR (300MHz, CDCl₃) δ 2.13 (p, 2H, J = 6.22Hz), 2.63 (t, 2H, J = 6.95Hz), 2.98 (t, 2H, J = 6.22Hz), 7.15 (m, 2H), 8.07 (m, 1H); ¹³C NMR δ 23.08, 29.88, 38.92, 116.74, 119.81, 121.56, 130.14, 132.58, 147.38, 152.52, 196.53.

Example 2: 6-Octyl-3,4-dihydro-2H-naphthalen-1-one (3).

[00159] 9-BBN (0.5 M solution in THF, 20.2 mL, 10.1 mmol) was added to 1-octene (1.6 mL, 10.1 mmol) at room temperature. The solution was stirred, at room temperature, overnight. After this time, K₃PO₄ (2.93 g, 13.8 mmol), Pd(Ph₃P)₄ (191 mg, 0.17 mmol, 1.8 mol %), KBr (1.2 g, 10.1 mmol) and degassed H₂O (0.18 mL, 10 mmol) were added. This was followed by a solution of compound **2** (2.7 g, 9.2 mmol) in dry degassed THF (10 mL). The reaction mixture was heated at 65 °C under argon for 2 hours. After cooling, the solution was acidified to pH 1 and extracted into EtOAc (100 mL). The aqueous layer was re-extracted with EtOAc (50 mL) and the combined organics washed with H₂O (20 mL) and brine (40 mL). The organic layer was dried over magnesium sulfate and concentrated under vacuum. The residue was purified by column chromatography (Silica gel, 5% EtOAc in hexanes) to provide 1.93 g of compound **3** (82%).

[00160] ^1H NMR (300MHz, CDCl_3) δ 0.85 (t, 3H, $J = 6.95\text{Hz}$), 1.24 (bs, 10H), 1.58 (p, 2H, $J = 6.95\text{Hz}$), 2.06 (p, 2H, $J = 5.85\text{Hz}$), 2.57 (t, 4H, $J = 6.95\text{Hz}$), 2.87 (t, 2H, $J = 6.22\text{Hz}$), 7.01 (s, 1H), 7.06 (d, 1H, $J = 8.05\text{ Hz}$), 7.91 (d, 1H, $J = 8.06\text{Hz}$); ^{13}C NMR δ 14.32, 22.88, 23.61, 29.44, 29.55, 29.66, 29.96, 31.32, 32.08, 36.31, 39.33, 127.12, 127.45, 128.73, 130.75, 144.70, 149.28, 198.09.

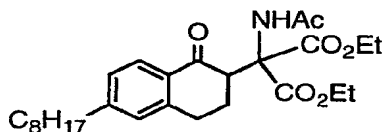
Example 3: 2-Bromo-6-octyl-3,4-dihydro-2H-naphthalen-1-one (4).



[00161] Cupric bromide (3.34 g, 15.0 mmol,) was heated at reflux in ethyl acetate (10 mL) with stirring. To this was added compound 3 (1.93 g, 7.5 mmol) in chloroform (10 mL). The reaction was heated at reflux for a further 6 hours and cooled. Copper bromide and cupric bromide residues were filtered off and the filtrate was decolorized with activated charcoal and filtered through a bed of Celite and washed with ethyl acetate (4x50 mL). The solvent was removed under reduced pressure and the residue was purified by column chromatography (Silica gel, 2% EtOAc in hexanes) to provide 2.02 g of compound 4 (80%).

[00162] ^1H NMR (300MHz, CDCl_3) δ 0.87 (t, 3H, $J = 6.95\text{Hz}$), 1.26 (bs, 10H), 1.61 (p, 2H, $J = 6.96\text{Hz}$), 2.46 (m, 2H), 2.62 (t, 2H, $J = 7.69\text{Hz}$), 2.86 (dt, 1H, $J = 16.34\text{Hz}$, 4.39Hz), 3.27 (dt, 1H, $J = 16.83\text{Hz}$, 4.39Hz), 4.69 (t, 1H, $J = 4.02\text{Hz}$), 7.07 (s, 1H), 7.14 (d, 1H, $J = 8.05\text{ Hz}$), 7.99 (d, 1H, $J = 8.05\text{Hz}$); ^{13}C NMR δ 14.34, 22.88, 26.42, 29.44, 29.57, 29.64, 31.25, 32.08, 32.32, 36.39, 127.75, 128.00, 128.73, 129.00, 144.30, 150.39, 190.54.

Example 4: 2-Acetylamino-2-(6-octyl-1-oxo-1,2,3,4-tetrahydro-naphthalen-2-yl)-malonic acid diethyl ester (5).

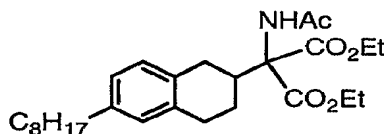


[00163] Sodium hydride (720 mg, 18.0 mmol) 60% in mineral oil was suspended in dry DMF (10 mL) and a solution of diethyl acetamidomalonate (3.26 g, 15 mmol) in dry DMF (10 mL) was added. The solution was stirred at 0 °C for 3 hours until the anion had formed. A solution of 4 (2.02 g, 6.0 mmol) in dry DMF (10 mL) was added and the solution warmed to room temperature and stirred overnight. The mixture was poured into

distilled water (50 mL), in an ice-bath, acidified to pH 3 with 1M hydrochloric acid and extracted with ethyl acetate (3×50 mL). The organic phases were washed with brine (2×30 mL) and dried over magnesium sulfate and concentrated under vacuum. The residue was purified by column chromatography (Silica gel, 40% EtOAc in hexanes) to provide 2.12 g of compound **5** (75%).

[00164] ^1H NMR (300MHz, CDCl_3) δ 0.85 (t, 3H, $J = 6.22\text{Hz}$), 1.24 (m, 16H), 1.58 (p, 2H, $J = 6.95\text{Hz}$), 1.97 (s, 3H), 2.59 (t, 2H, $J = 7.32\text{Hz}$), 2.83-3.21 (m, 4H), 3.88 (dd, 1H, $J = 14.00\text{Hz}$, 3.68Hz), 4.14-4.32 (m, 4H), 6.86 (s, 1H), 7.03 (s, 1H), 7.07 (d, 1H, $J = 8.69\text{Hz}$), 7.84 (d, 1H, $J = 8.36\text{Hz}$); ^{13}C NMR δ 14.05, 14.16, 14.30, 22.85, 23.31, 26.98, 29.40, 29.49, 29.61, 29.98, 31.28, 32.05, 36.32, 56.16, 62.40, 63.13, 66.33, 127.16, 127.63, 128.78, 144.84, 150.07, 166.38, 168.70, 169.83, 197.63.

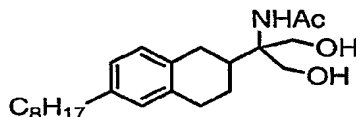
Example 5: 2-Acetylamino-2-(6-octyl-1,2,3,4-tetrahydro-naphthalen-2-yl)-malonic acid diethyl ester (**6**).



[00165] To a solution of triethylsilane (1.3 mL, 8.2 mmol) in 5 mL of CH_2Cl_2 was added compound **5** (1 g, 2.1 mmol) in 5 mL of CH_2Cl_2 . The reaction mixture was stirred at room temperature under Ar and TiCl_4 (0.09 mL, 8.2 mmol) was added dropwise. The resulting solution was stirred for 12 hours, cooled to 0°C and quenched by slow addition of 10 mL of saturated NaHCO_3 . The aqueous layer was extracted with CH_2Cl_2 (3×30 mL). The combined organic layers were washed with brine (2×30 mL) and dried over magnesium sulfate and concentrated under vacuum. The residue was purified by column chromatography (Silica gel, 20% EtOAc in hexanes) to provide 630 mg of compound **6** (65%).

[00166] ^1H NMR (300MHz, CDCl_3) δ 0.87 (t, 3H, $J = 6.46\text{Hz}$), 1.26 (m, 16H), 1.58 (p, 2H, $J = 6.79\text{Hz}$), 2.03 (s, 3H), 2.28 (b, 1H), 2.49-2.68 (m, 4H), 2.82-2.92 (m, 2H), 4.20-4.34 (m, 4H), 6.69 (s, 1H), 6.89-7.05 (m, 3H); ^{13}C NMR δ 14.21, 14.25, 14.37, 22.92, 23.37, 25.48, 29.50, 29.63, 29.72, 29.76, 30.39, 31.93, 32.13, 35.78, 40.33, 62.46, 62.79, 68.80, 126.04, 128.81, 129.30, 132.85, 136.28, 140.66, 150.07, 167.63, 168.32, 169.42.

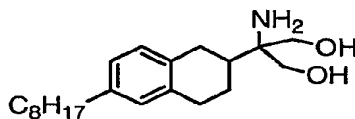
Example 6: N-[2-Hydroxy-1-hydroxymethyl-1-(6-octyl-1,2,3,4-tetrahydro-naphthalen-2-yl)-ethyl]-acetamide (7).



[00167] Lithium borohydride (2M solution in THF, 0.88 ml, 1.76 mmol) was added to compound **6** (200 mg, 0.44 mmol) in 5 ml THF at 0°C. The reaction mixture was stirred at room temperature for 48 hours and diluted with 40 ml ethyl acetate. The solution was washed with brine (2×20 mL) and dried over magnesium sulfate and concentrated under vacuum. The residue was purified by column chromatography (Silica gel, 4% MeOH in CH₂Cl₂) to provide 55 of mg compound **7** (33%).

[00168] ¹H NMR (300MHz, CDCl₃) δ 0.88 (t, 3H, J = 6.56Hz), 1.29 (m, 10H), 1.57 (p, 2H, J = 6.25Hz), 1.94-1.98 (m, 2H), 2.05 (s, 3H), 2.33 (m, 1H), 2.51 (t, 2H, J = 7.32), 2.60-2.85 (m, 4H), 3.69 (d, 2H, J = 11.61), 3.89 (dd, 2H, J = 11.61Hz, 7.25Hz), 6.22 (s, 1H), 6.88-6.99 (m, 3H); ¹³C NMR δ 14.38, 22.92, 24.20, 24.35, 29.52, 29.66, 29.73, 29.95, 30.32, 31.94, 32.14, 35.78, 38.26, 63.55, 64.34, 64.46, 126.18, 128.85, 129.30, 133.06, 136.22, 140.75, 172.40.

Example 7: 2-Amino-2-(6-octyl-1,2,3,4-tetrahydro-naphthalen-2-yl)-propane-1,3-diol (VPC104061).

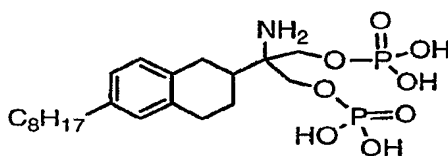


[00169] A solution of compound **7** (53 mg, 0.14 mmol) and LiOH·H₂O (45 mg, 1.1 mmol) in MeOH (3 ml), THF (1.5 ml) and water (3 ml) was stirred at 50 °C for 5 hours and diluted with ethyl acetate (20 ml). The solution was washed with brine (2×10 mL) and dried over magnesium sulfate and concentrated under vacuum. The residue was purified by column chromatography (Silica gel, 50% MeOH in CH₂Cl₂) to provide 35 mg of compound VPC104061 (75%).

[00170] ¹H NMR (300MHz, CDCl₃) δ 0.88 (t, 3H, J = 6.17Hz), 1.29 (m, 10H), 1.56 (p, 2H, J = 6.17Hz), 1.82-1.98 (m, 2H), 2.51 (t, 2H, J = 6.95), 2.58-2.88 (m, 5H), 3.19 (b, 4H), 3.61 (d, 2H, J = 10.98), 3.73 (d, 2H, J = 10.61Hz), 6.87-6.98 (m, 3H); ¹³C NMR δ 14.37, 22.93, 24.02, 29.32, 29.53, 29.70, 29.75, 29.85, 30.26, 31.94, 32.14, 35.08, 39.58,

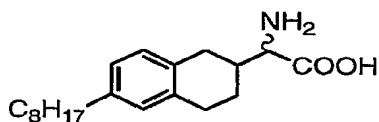
57.74, 66.13, 66.19, 126.09, 128.81, 129.39, 133.28, 136.26, 140.64. MS (ESI) m/z 334.1 $[M+H]^+$.

Example 8: Phosphoric acid mono -[2-amino -2-(6-octyl -1,2,3,4- tetrahydro-naphthalen-2-yl)- 3- phosphonooxypropyl] ester (VPC104081).



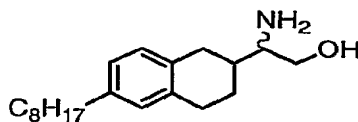
[00171] Phosphorus pentoxide (2.0 g, 14 mmol) in phosphoric acid (85% in water, 2 ml, 29 mmol) was added to VPC104061 (25 mg, 0.07 mmol). The mixture was stirred at 100°C for 2 hours and cooled to 0°C. The product was precipitated by adding water (14 mg, 37%). MS (ESI) m/z 494.4 $[M+H]^+$.

Example 9: Amino-(6-octyl-1,2,3,4-tetrahydro-naphthalen-2-yl)-acetic acid (8).



[00172] Compound **6** (300 mg, 0.65 mmol) was added to 12 M HCl (10 ml). The mixture was heated to reflux and MeOH (5 ml) was added until the mixture became homogenous. Reflux was continued for 2 hours until the starting material was consumed as determined by thin layer chromatography (TLC). The reaction mixture was concentrated under reduced pressure and co-evaporated with MeOH and diethyl ether multiple times. The desired compound **8** was recrystallized from diethyl ether and hexanes to provide a light brown solid and used directly for the next reaction.

Example 10: 2-Amino-2-(6-octyl-1,2,3,4-tetrahydro-naphthalen-2-yl)-ethanol (VPC104059).

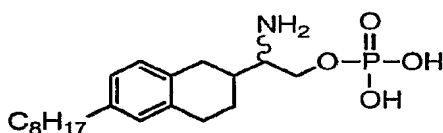


[00173] The amino acid **8** prepared in Example 9 was added to a refluxing solution of lithium aluminum hydride (62 mg, 1.63 mmol) in THF (10 ml). The reaction mixture was heated at reflux for 12 hours, subsequently cooled to 0°C and 10 M NaOH was added and stirred for 20 minutes. Ethyl acetate (20 ml) was added and the mixture was filtered through Celite and magnesium sulfate. The filtrate was concentrated under vacuum and

purified by column chromatography (Silica gel, 50% MeOH in CH₂Cl₂) to provide 41 mg of the product, VPC104059 (21%, two steps).

[00174] ¹H NMR (300MHz, CDCl₃) δ 0.88 (t, 3H, J = 6.39Hz), 1.28 (m, 10H), 1.55 (p, 2H, J = 7.16Hz), 1.67-2.11 (m, 3H), 2.48 (t, 2H, J = 7.69), 2.56-2.83 (m, 5H), 3.19 (b, 4H), 3.47-3.75 (m, 2H), 6.82-6.96 (m, 3H); ¹³C NMR δ 13.34, 22.58, 26.45, 29.14, 29.21, 29.29, 29.47, 31.70 31.90, 32.26, 35.40, 47.56, 125.64, 128.36, 128.86, 128.93, 133.19, 139.93, MS (ESI) m/z 303.9 [M+H]⁺.

Example 11: Phosphoric acid mono-[2-amino-2-(6-octyl-1,2,3,4-tetrahydro-naphthalen-2-yl)-ethyl] ester (VPC104127).

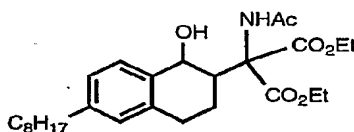


[00175] Phosphorus pentoxide (1.5 g, 10.5 mmol) in phosphoric acid (85% in water, 1.5 ml, 22 mmol) was added to VPC104059 (25 mg, 0.08 mmol). The mixture was stirred at 100°C for 2h and cooled to 0°C. The product was precipitated by adding water (10 mg, 50%). MS (ESI) m/z 384.2 [M+H]⁺.

Example 12: Synthesis of structure (X)

[00176] The synthesis of an ether containing compound having formula IX is illustrated in Scheme 2 (Fig. 3). Keto-alcohol **1A**, is converted to the keto-ether **1B**, using standard reagents and techniques. The keto-ether is halogenated to provide halo-ether **1C** in a manner similar to Example 3. The halo-ether is alkylated to provide diester-ether **1D**, in a manner similar to the procedure described in Example 4. The diester is converted to ether-triol **1E**, using standard reducing agents known in the art. The triol is converted to a diol and deprotected using standard methods known in the art, to provide compound **IX**.

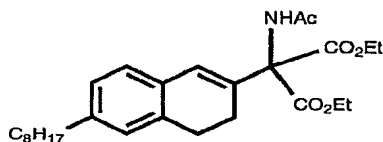
Example 13: 2-Acetylamino-2-(6-octyl-1-hydroxy-1,2,3,4-tetrahydro-naphthalen-2-yl)-malonic acid diethyl ester (A).



[00177] To a solution of sodium borohydride (75 mg, 2.00 mmol) in 5 ml of ethanol at room temperature is added 2-acetylamino-2-(6-octyl-1-oxo-1,2,3,4-tetrahydro-naphthalen-2-yl)-malonic acid diethyl ester (compound **5**) (1.00 g, 2.1 mmol) in 5 ml of ethanol. The

reaction mixture is stirred at room temperature under argon for an additional hour, quenched by the addition of water (20 mL) and methylene chloride (20 mL). The organic layer is removed and the aqueous layer is extracted with methylene chloride (2 X 20 mL). The combined organic layers were washed with brine (2 X 20 mL) and dried over magnesium sulfate and concentrated under vacuum. The residue is purified by column chromatography (silica gel, 20% ethyl acetate in hexanes) to provide 755 mg of compound A (75%).

Example 14: 2-Acetylamino-2-(6-octyl-3,4-dihydro-naphthalen-2-yl)-malonic acid diethyl ester (B)



[00178] 2-Acetylamino-2-(6-octyl-1-hydroxy-1,2,3,4-tetrahydro-naphthalen-2-yl)-malonic acid diethyl ester (compound A, 755 mg, 1.58 mmol) is dissolved in acetic anhydride (5 mL) followed at 0°C by a catalytic amount of ferric chloride (66 mg, 0.4 mmol). The reaction is stirred at 0°C for an additional 2 hours, and 20 mL of diethyl ether is added. The reaction is carefully poured into 50 mL of ice cold water and the organic layer quickly separated. The organic layer is re-extracted with methylene chloride (2 X 20 mL) and the combined organic layers were washed once with brine (20 mL) and dried over magnesium sulfate. The organic layer is concentrated under vacuum and the residue is purified by column chromatography (silica gel, 10% ethyl acetate in hexanes) to provide 458 mg of compound B (60%). The complete synthesis is illustrated in Fig. 4.

Example 14: Sphingosine Kinase Assay

[00179] Recombinant sphingosine kinase type 2 (SPHK2) is prepared by forcing the expression of the mouse or human recombinant enzyme by transfecting the relevant plasmid DNA into HEK293T cells. After about 60 hours, cells are harvested, broken and the non-microsomal (e.g., soluble) fraction is retained. The broken cell supernatant fluid containing the recombinant enzyme is mixed with test compounds (FTY720, AA151, VIII and XVIII) (5 – 50 micromolar) and γ -32P-ATP and incubated for 0.5 – 2.0 hours at 37°C. The lipids in the reaction mixture are extracted into an organic solvent and displayed by normal phase thin layer chromatography. The radio-labeled bands are detected by

autoradiography, scraped from the plate and quantified by scintillation counting. In the histogram shown, (Figs. 5A, 5B and 6) sphingosine was present at 15 μ M, and the test compounds were at a concentration of 50 μ M, incubation time was 20 minutes.

Example 15: GTP γ S-35 binding Assay

[00180] This assay illustrates agonist activation of G protein coupled receptors (GPCRs) in isolation. The assay forces expression concomitantly of a recombinant GPCR (*e.g.*, the S1P1-5 receptor) and each of the three subunits (typically, α -2, β -1, or γ -2) of a heterotrimeric G protein in a HEK293T cell by transfecting the cell with four plasmid DNAs encoding the respective proteins. About 60 hours after transfection the cells are harvested, opened, and the nucleus discarded. The crude microsome is prepared from the remainder. Agonist (*e.g.*, S1P) stimulation of the receptor-G protein complex on the microsomes results in the exchange of GTP for GDP on the α -subunit in a dose-dependent manner. The GTP-bound α -subunit is detected using a GTP analog (GTP γ S-35), which is a radionuclide (sulfur-35) labeled phosphothionate that is not hydrolyzed to GDP. The microsomes with the adherent G proteins are collected by filtration and the bound GTP γ S-35 quantified in a liquid scintillation counter. The assay yields relative potency (EC₅₀ values) and maximum effect (efficacy, E_{max}). Antagonist activity is detected as rightward shifts in the agonist dose-response curve in the presence of a fixed amount of antagonist. If the antagonist behaves competitively, the affinity of the receptor/antagonist pair (K_i) can be determined.

[00181] The phosphorylated forms of compound VIII (a mixture of mono, XXVI, and diphosphorylated, XXVII, isomers) and compound XXVIII are low potency, partial agonists at the S1P₃ receptor (See Fig. 8.). Compounds XXVI, XXVII and XXVIII are more potent at S1P₁ and less potent at S1P₃, relative to S1P. (See Fig. 7.) The assay was performed as described in Davis, M.D., J.J. Clemens, T.L. Macdonald and K.R. Lynch (2005) "S1P Analogs as Receptor Antagonists" Journal of Biological Chemistry, vol. 280, pp. 9833-9841.

Example 16: Lymphopenia Assay

[00182] Compounds (*e.g.*, primary alcohols such as compound VIII) are dissolved in 2% hydroxypropyl beta-cyclodextrin and introduced into groups of mice by oral gavage at doses from .01, 1.0 and 10 mg/kg body weight. After 24 hours and 48 hours, the mice are lightly anesthetized and *ca.* 0.1 ml of blood is drawn from the orbital sinus. The number

of lymphocytes (in thousands per microliter of blood; normal is 4-11) is determined using a Hemavet blood analyzer. In the histogram showing compound VIII, the 100% value for the vehicle treated mouse was 7.5 at 24 hours; at 96 hours it was 5. There were three mice/group, the strain was mixed sv129 x C57BL/6. Active compounds (*e.g.*, compound VIII and compound XXVIII) are dissolved in acidified DMSO at 20 mM, and diluted 1:20 into 2% hydroxypropyl beta-cyclodextrin in water with mixing. This solution is introduced into mice by intraperitoneal (i.p.) injection at doses of 0.01, 1.0 and 10 mg/kg body weight.

[00183] In Fig. 9 the results of an assay using compound VIII and compound XXVIII administered via oral gavage are graphically illustrated. The total lymphocyte counts (k/ μ l) 24 hours after an IV dose of compound VIII is reported.

[00184] Compounds were dissolved in 2% hydroxypropyl β -cyclodextrin and administered to sets of three, 12 week old C57Bl/6 female mice by oral gavage (PO). At 18 hours post dosing, blood was drawn from the orbital sinus and lymphocyte numbers were determined with a Hemavet blood analyzer. Doses were: compound XXIX (1 mg/kg), compound VIII (10 mg/kg), and FTY720 (0.3 mg/kg). Compound XXIX, 3,5-diphenyl-1,2,4-oxadiazole, is a S1P1 receptor agonist described by Li et al. (Journal Medicinal Chemistry, vol. 48, p. 6169 (2005)) was used as a comparison. The results are illustrated in Fig. 10.

Example 17: Heart Rate Assay

Mice were dosed with compound VIII, FTY720 (intravenous, 3 mg/kg) or vehicle (2% hydroxypropyl beta-cyclodextrin) and heart rate measured at 1 hour post dosing. Heart rate was captured in unrestrained, conscious animals using the ECGenie™ system. The results are illustrated in Fig. 11.

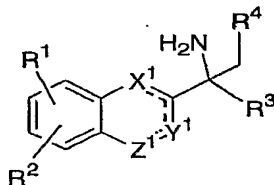
[00185] The invention should not be construed to be limited solely to the assays and methods described herein, but should be construed to include other methods and assays as well. Other methods which were used but not described herein are well known and within the competence of one of ordinary skill in the art of chemistry, biochemistry, molecular biology, and clinical medicine. One of ordinary skill in the art will know that other assays and methods are available to perform the procedures described herein.

[00186] The abbreviations used herein have their conventional meaning within the clinical, chemical, and biological arts. In the case of any inconsistencies, the present disclosure, including any definitions therein will prevail.

[00187] The disclosures of each and every patent, patent application, and publication cited herein are expressly incorporated herein by reference in their entirety into this disclosure. Illustrative embodiments of this disclosure are discussed and reference has been made to possible variations within the scope of this disclosure. These and other variations and modifications in the disclosure will be apparent to those skilled in the art without departing from the scope of the disclosure, and it should be understood that this disclosure and the claims shown below are not limited to the illustrative embodiments set forth herein.

We Claim:

1. A compound of formula I:



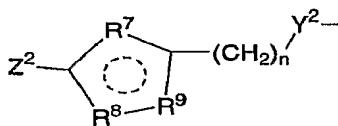
(I)

wherein

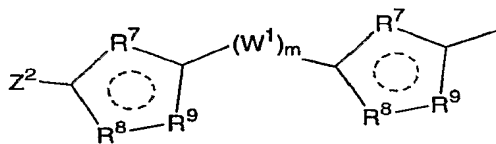
X^1 , Y^1 and Z^1 are independently O, CR^a , CR^aR^b , N, NR^c , or S;

R^1 is hydrogen, halo, (C_1-C_{10}) alkyl, (C_1-C_{10}) haloalkyl, or (C_1-C_{10}) alkoxy;

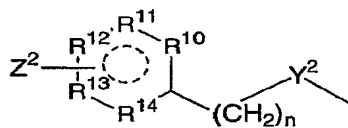
R^2 is hydrogen, halo, (C_1-C_{20}) alkyl, (C_1-C_{20}) alkoxy; (C_2-C_{26}) alkoxyalkyl; (C_2-C_{20}) alkenyl, (C_2-C_{20}) alkynyl, (C_3-C_{12}) cycloalkyl, (C_6-C_{10}) aryl, (C_7-C_{30}) arylalkyl, (C_2-C_{10}) heterocyclic, and (C_5-C_{10}) heteroaryl; or R^2 can be a group having formula II, III, IV, V, or VI;



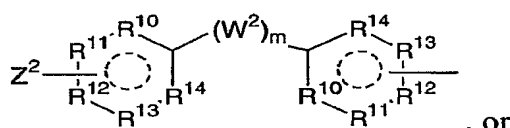
II



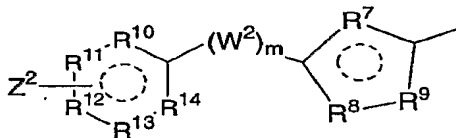
III



IV



V



VI

where R^7 , R^8 , R^9 , R^{10} , R^{11} , R^{12} , R^{13} , and R^{14} are independently O, S, C, CR^{15} , $CR^{16}R^{17}$, C=O, N or NR^{18} ;

R^{15} , R^{16} and R^{17} are independently hydrogen, halo, (C_1-C_{10}) alkyl, (C_1-C_{10}) alkyl substituted with halo, hydroxy, (C_1-C_{10}) alkoxy, or cyano; and where R^{18} can be hydrogen or (C_1-C_{10}) alkyl;

where at least one of R^{10} , R^{11} , R^{12} , R^{13} , or R^{14} is a heteroatom;

where Z^2 is (C_1-C_6) alkyl, (C_3-C_8) cycloalkyl, substituted alkyl, (C_2-C_6) alkenyl,

(C₂-C₆)alkynyl, aryl, alkyl substituted aryl, arylalkyl, or aryl substituted arylalkyl; wherein the alkyl groups of Z² are optionally substituted with 1, 2, 3, or 4 substituent groups where the substituent groups independently are halo, (C₁-C₁₀)alkoxy or cyano;

○ indicates one or more optional double bonds;

wherein Y² is O, C=O, or CH₂; W¹ is a bond or -CH₂-CH₂-CH₂-; W² is a bond or -CH₂- and m is 1, 2, or 3, or (C=O)(CH₂)₁₋₅ and m is 1; and n is 0, 1, 2, or 3;

each --- represents an optional double bond; R³ is hydrogen, (C₁-C₁₀)alkyl, or (C₁-C₁₀)alkoxy;

R⁴ is hydroxyl (-OH), phosphate (-OPO₃H₂), phosphonate (-CH₂PO₃H₂), or *alpha*-substituted phosphonate;

R^a, R^b, and R^c are independently hydrogen, or (C₁-C₁₀)alkyl;

wherein the alkyl groups of R¹ are optionally substituted with 1, 2, 3, or 4 substituent groups where the substituent groups independently are halo, (C₁-C₁₀)alkoxy or cyano;

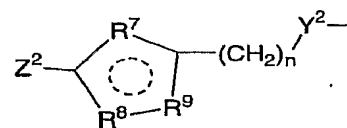
wherein any of the alkyl, alkenyl, alkynyl, cycloalkyl, aryl, heterocyclic, or heteroaryl groups of R² are optionally substituted with 1, 2, 3, or 4 substituent groups where the substituent groups independently are oxo (=O), imino (=NR^d), (C₁-C₁₀)alkyl, (C₁-C₁₀)alkoxy, or C₆-aryl; or wherein one or more of the carbon atoms in the R² alkyl groups can be independently replaced with non-peroxide oxygen, sulfur or NR^c; the alkyl groups of R³ are optionally substituted with 1, or 2 hydroxy groups; and R^d is hydrogen, or (C₁-C₁₀)alkyl; or

a pharmaceutically acceptable salt or ester thereof.

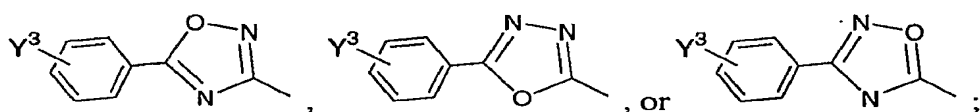
2. The compound of claim 1 wherein R¹ is hydrogen, fluorine, chlorine, bromine, trifluoromethyl, methoxy, (C₁-C₆)alkyl, (C₁-C₆)haloalkyl, (C₁-C₆)alkyl substituted with, alkoxy or cyano, alkyl-substituted aryl, aryl-substituted alkyl, or aryl-substituted arylalkyl.
3. The compound of claim 2 wherein R¹ is hydrogen, trifluoromethyl, or -CH₂CF₃.
4. The compound of claim 2 wherein R¹ is benzyl, phenylethyl, or methyl benzyl.

5. The compound of any of claims 1-4 wherein R^2 comprises
 $-\text{CH}_2-\text{CH}_2-\text{O}-\text{CH}_2-\text{CH}_2-\text{O}-$.

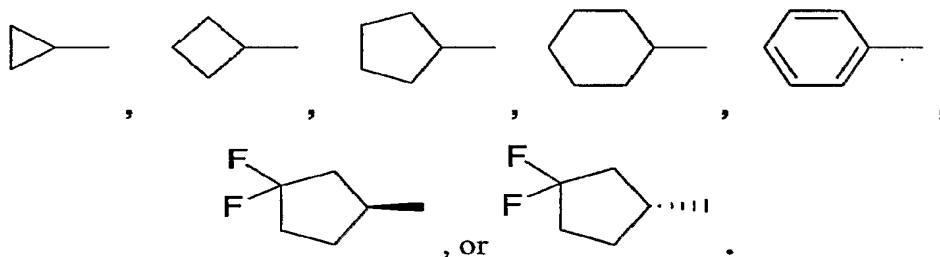
6. The compound of any of claims 1-4 wherein R^2 is



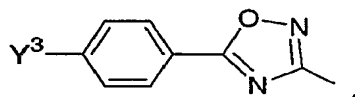
7. The compound of claim 6 wherein R^2 is:



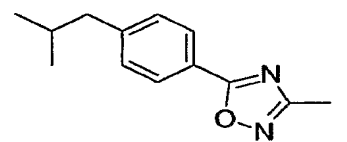
where Y^3 is $(\text{CH}_3)_3\text{C}-$, $\text{CH}_3\text{CH}_2(\text{CH}_3)_2\text{C}-$, $\text{CH}_3\text{CH}_2\text{CH}_2-$,
 $\text{CH}_3(\text{CH}_2)_2\text{CH}_2-$, $\text{CH}_3(\text{CH}_2)_4\text{CH}_2-$, $(\text{CH}_3)_2\text{CHCH}_2-$, $(\text{CH}_3)_3\text{CCH}_2-$,
 $\text{CH}_3\text{CH}_2\text{O}-$, $(\text{CH}_3)_2\text{CHO}-$, or $\text{CF}_3\text{CH}_2\text{CH}_2-$ or a group having the formula:.



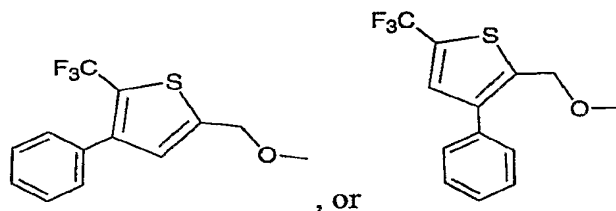
8. The compound of claim 7 wherein R^2 is:



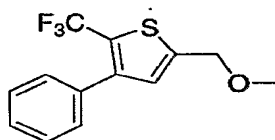
9. The compound of claim 8 wherein R^2 is:



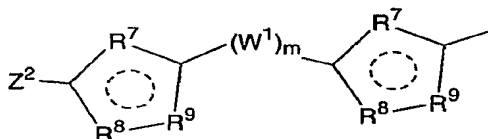
10. The compound of claim 6 wherein R^2 is:



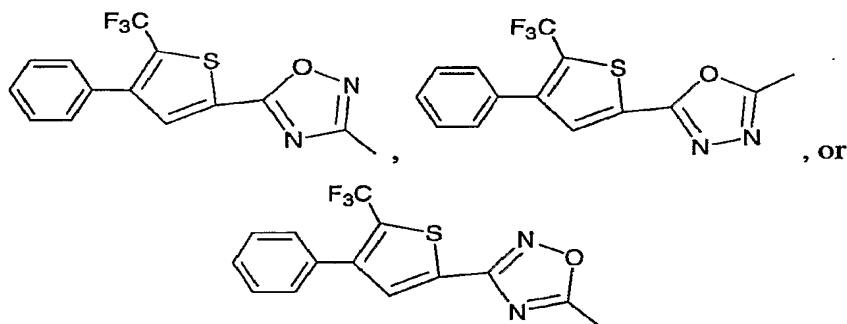
11. The compound of claim 10 wherein R^2 is



12. The compound of any of claims 1-4 wherein R^2 has formula IV

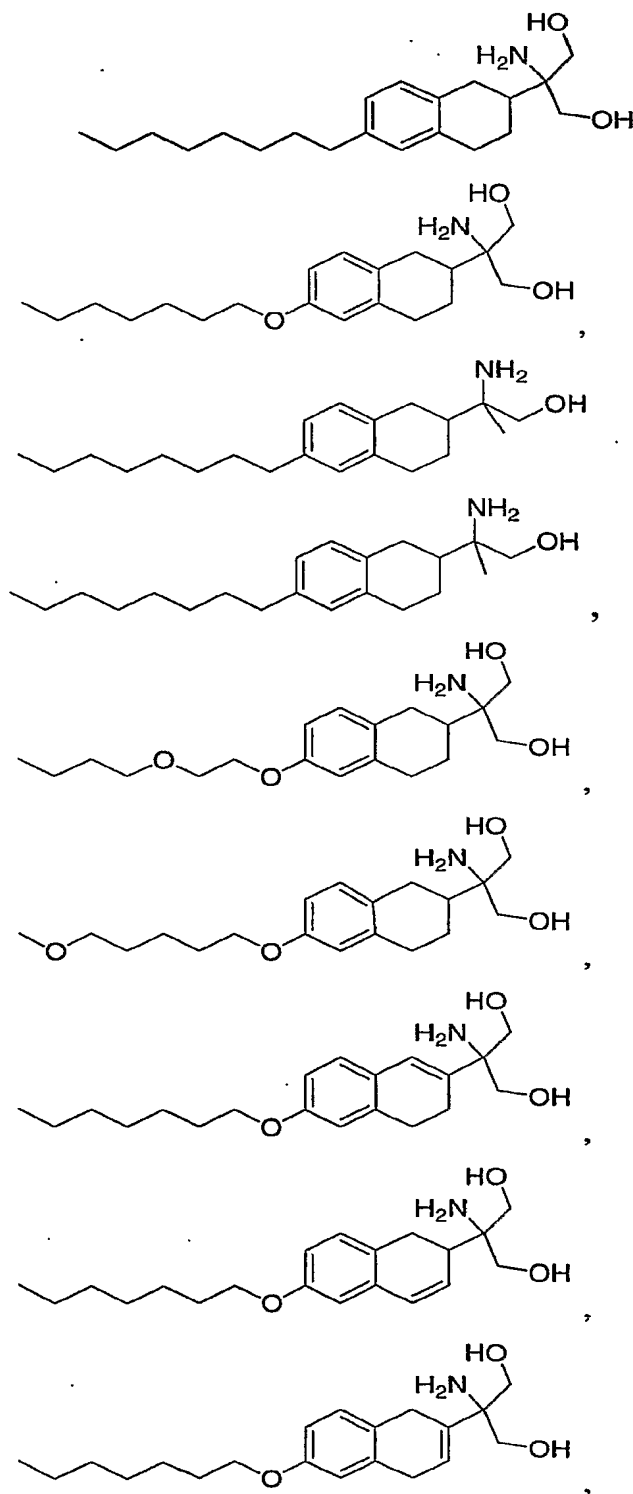


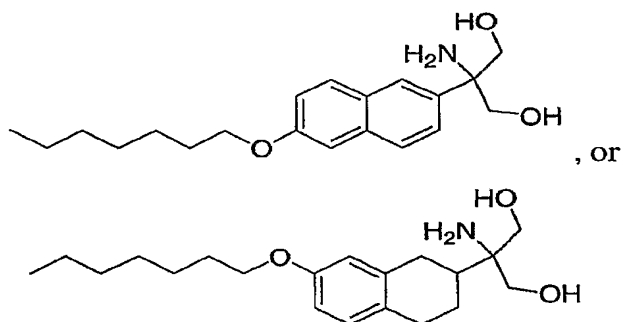
13. The compound of claim 12 wherein R^2 is



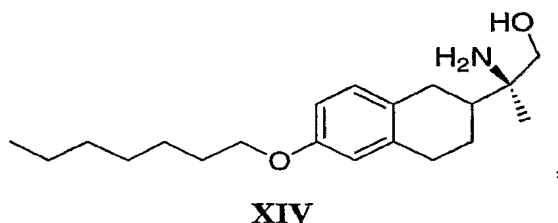
14. The compound of any of claims 1-4 wherein R^2 is (C_1-C_{10}) alkyl, (C_2-C_{10}) alkenyl and (C_2-C_{14}) alkynyl, (C_1-C_{10}) alkoxy or (C_2-C_{16}) alkoxyalkyl.
15. The compound of claim 14 wherein R^2 is (C_1-C_{10}) alkyl, (C_1-C_{10}) alkoxy or (C_2-C_{12}) alkoxyalkyl.
16. The compound of claim 15 wherein R^2 is methyl, ethyl, propyl, butyl, pentyl, hexyl, heptyl, octyl, trifluoromethyl, trifluoroethyl, trifluoromethoxy, trifluoroethoxy, methoxy, ethoxy, propoxy, butoxy, pentoxy, heptoxy, or octoxy.
17. The compound of any of claims 1-16 wherein each of X^1 , Y^1 and Z^1 is CH_2 .
18. The compound of any of claims 1-17 wherein R^3 is hydrogen, methyl, hydroxymethyl, ethyl, hydroxyethyl, propyl, or isopropyl.

19. The compound of claim 18 wherein R^3 is hydrogen, methyl, hydroxymethyl, ethyl, or hydroxyethyl.
20. The compound of any of claims 1-19 having the formula





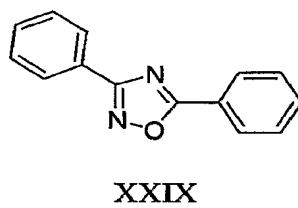
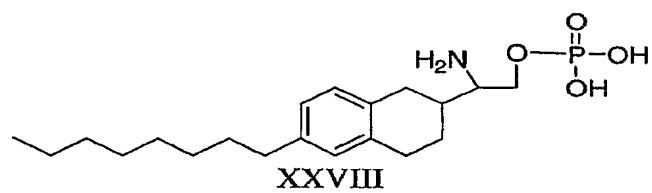
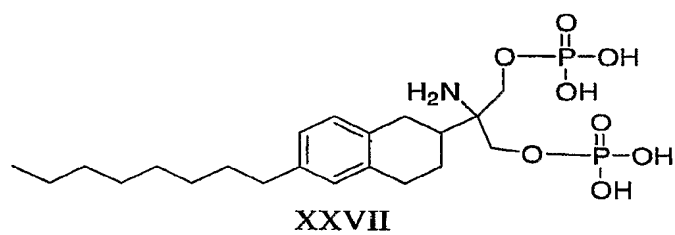
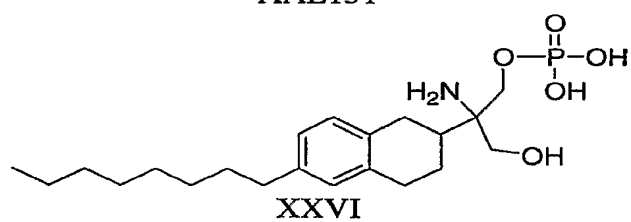
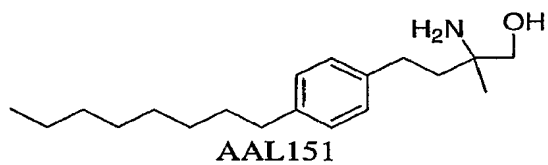
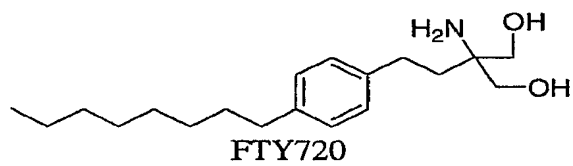
21. The compound of claim 20 having the formula:



22. A method for prevention or treatment of a pathological condition or symptom in a mammal, wherein the activity of sphingosine 1-phosphate receptors is implicated and agonism of such activity is desired, comprising administering to said mammal an effective amount of a compound of any of claims 1-21.
23. The method of statement 21, wherein the pathological condition is an autoimmune disease.
24. The method of claim 22, wherein the autoimmune disease is uveitis, type I diabetes, rheumatoid arthritis, inflammatory bowel diseases, or multiple sclerosis.
25. The method of claim 24, wherein the autoimmune disease is multiple sclerosis.
26. The method of claim 25, wherein the pathological condition is altering lymphocyte trafficking.
27. The method of claim 26, wherein the treatment is altering of lymphocyte trafficking.
28. The method of claim 27, wherein lymphocyte trafficking provides prolonged allograft survival.
29. The method of claim 28, wherein the allograft is for transplantation.
30. A method for prevention or treatment of a pathological condition or symptom in a mammal, wherein the activity S1P lyase implicated and inhibition of the S1P lyase is desired, comprising administering to said mammal an effective amount of a compound of any of claims 1-21.

31. The use of a compound of any of claims 1-21 for use in medical therapy.
32. Use of a compound of any of claims 1-21, to prepare a medicament useful for prevention or treatment of a pathological condition or symptom in a mammal, wherein the activity of sphingosine 1-phosphate receptors is implicated.
33. The use of claim 32, wherein the medicament comprises a liquid carrier.

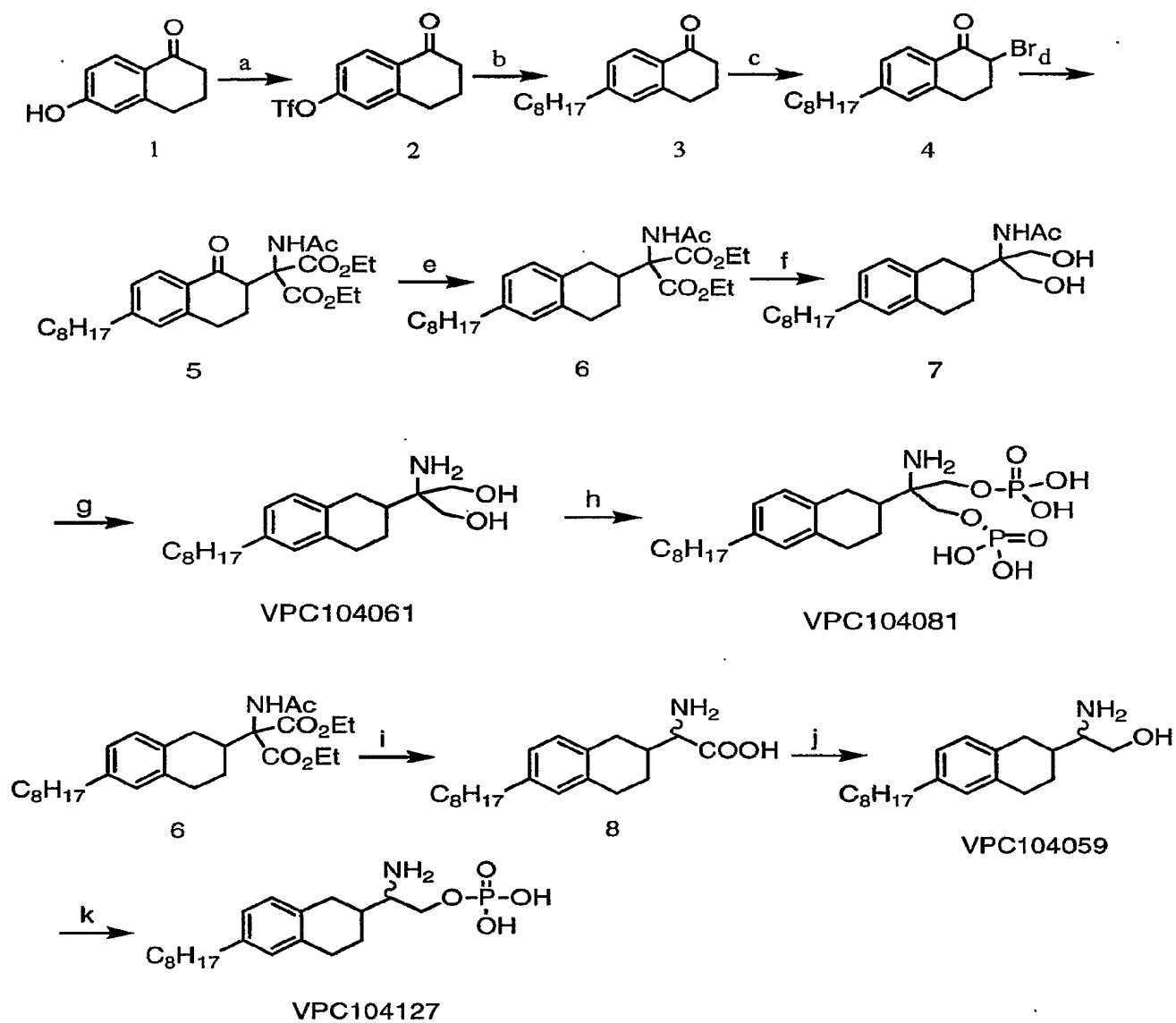
1/11

Fig. 1

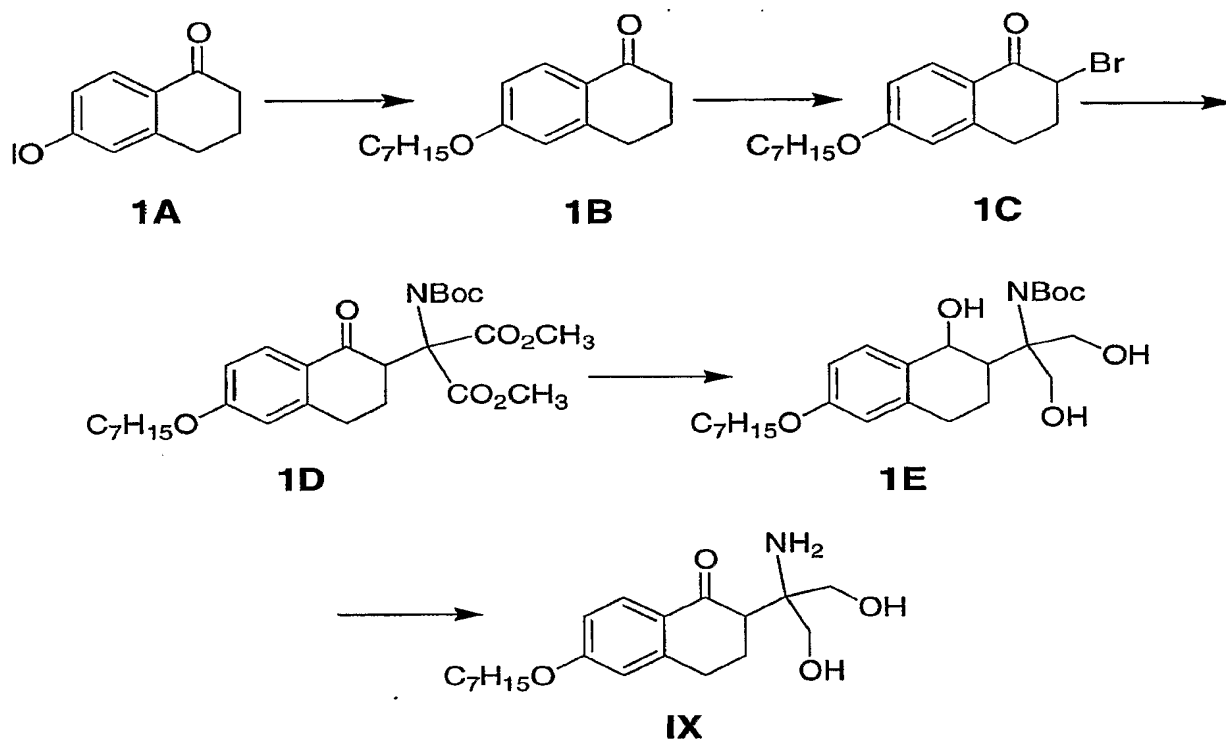
2/11

Fig. 2

Scheme 1



3/11

Fig. 3Scheme 2

4/11

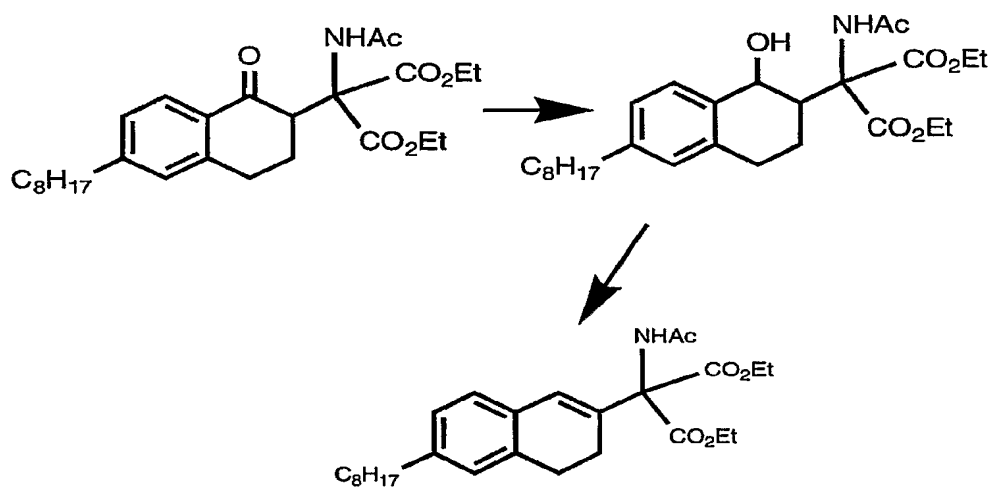
Fig. 4Scheme 3

Fig. 5B

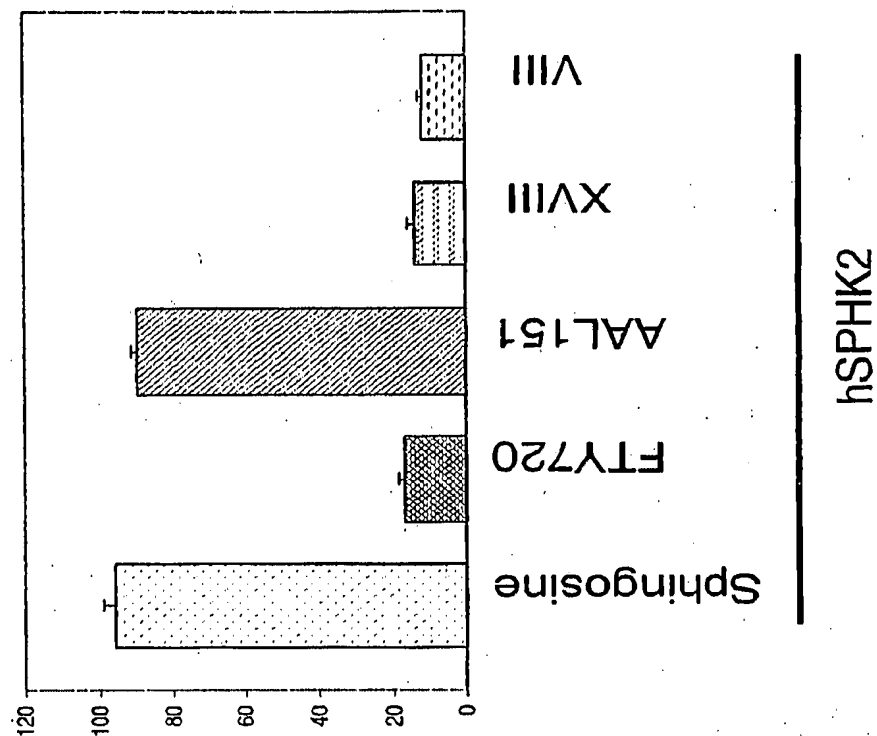


Fig. 5A

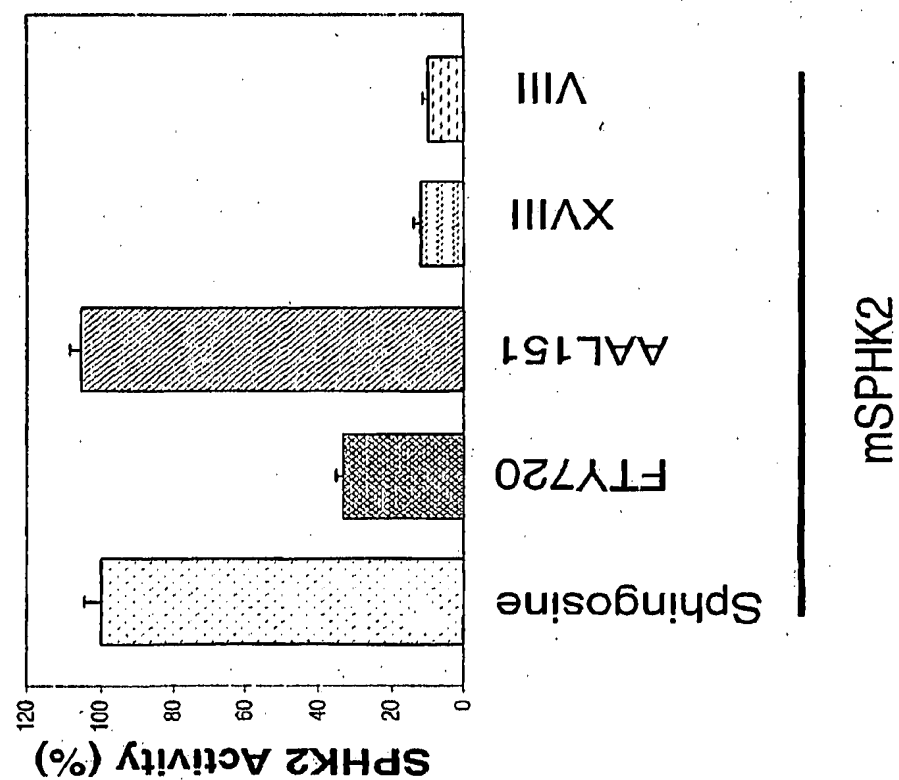


Fig. 6

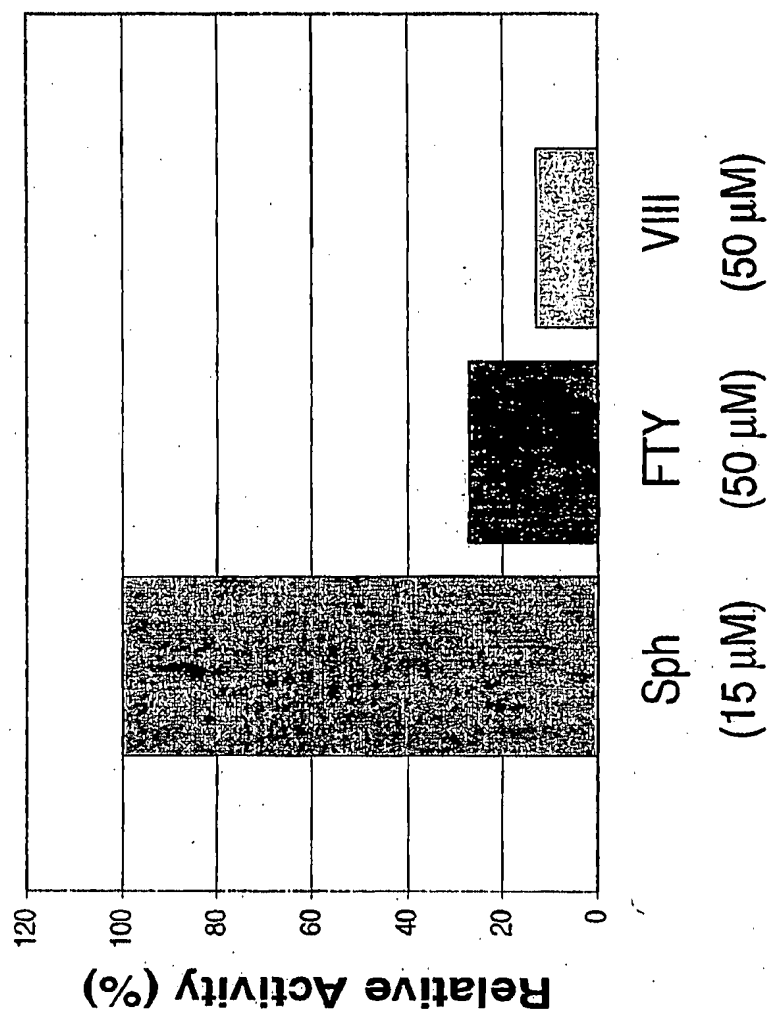


Fig. 7

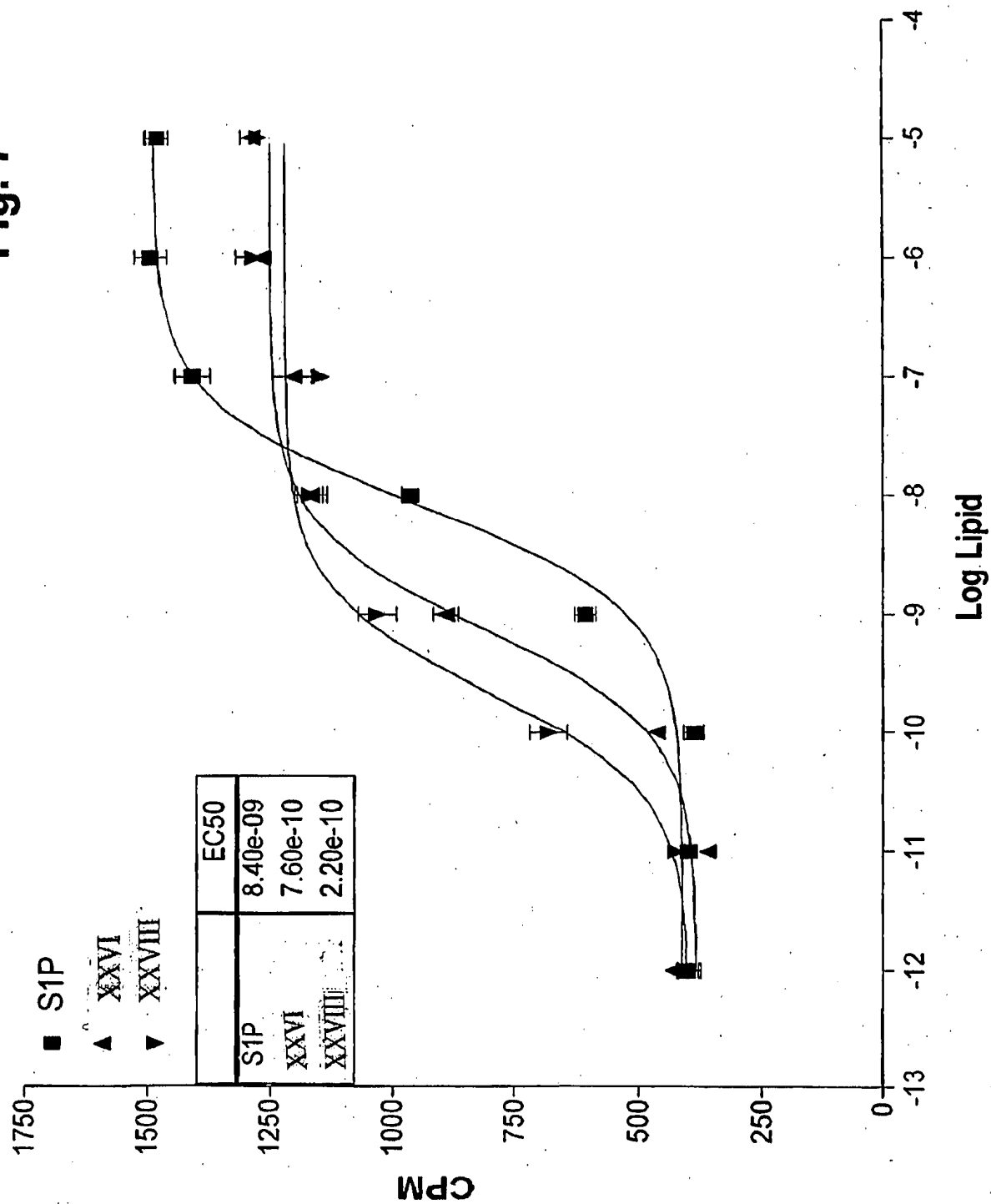


Fig. 8

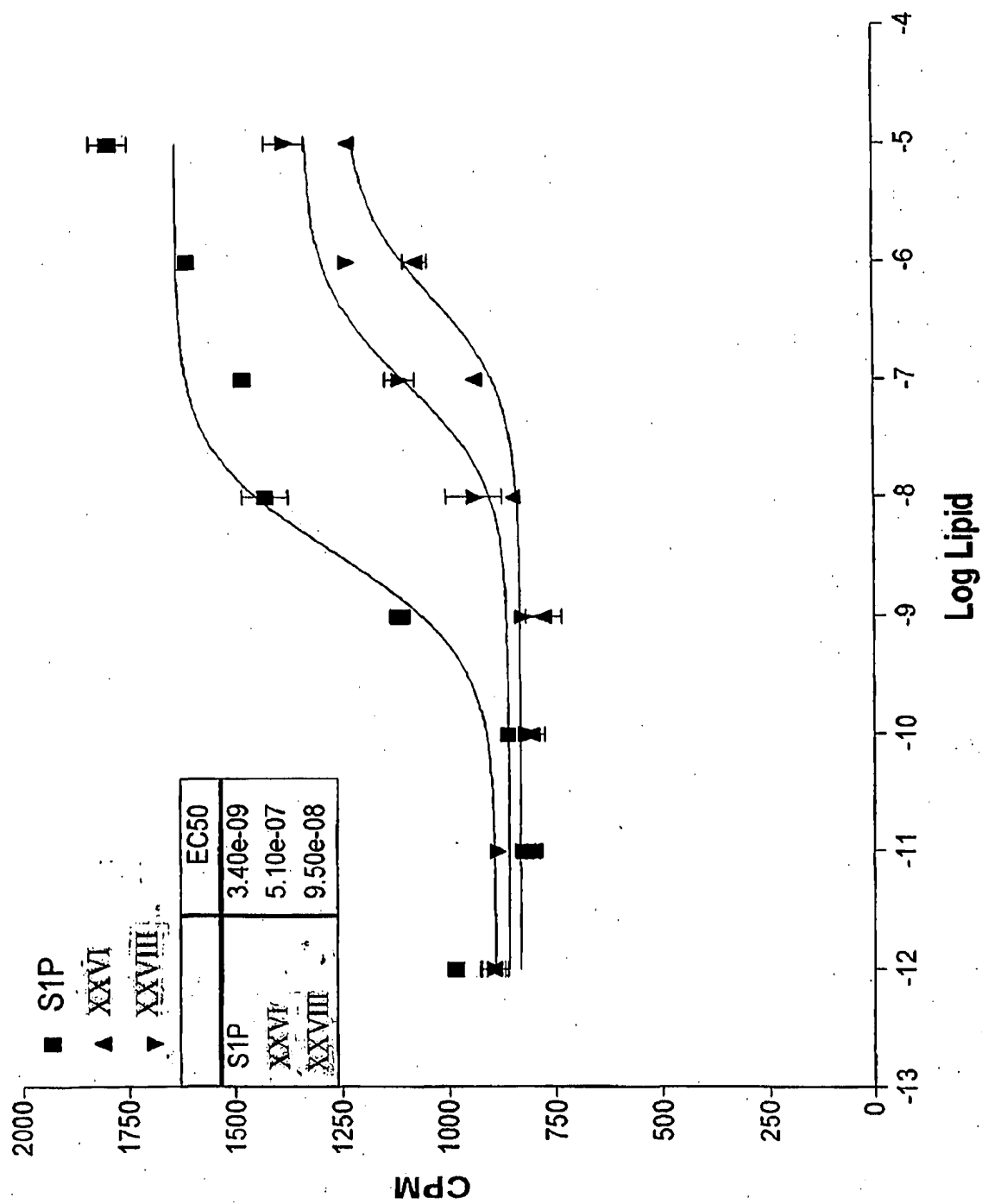


Fig. 9

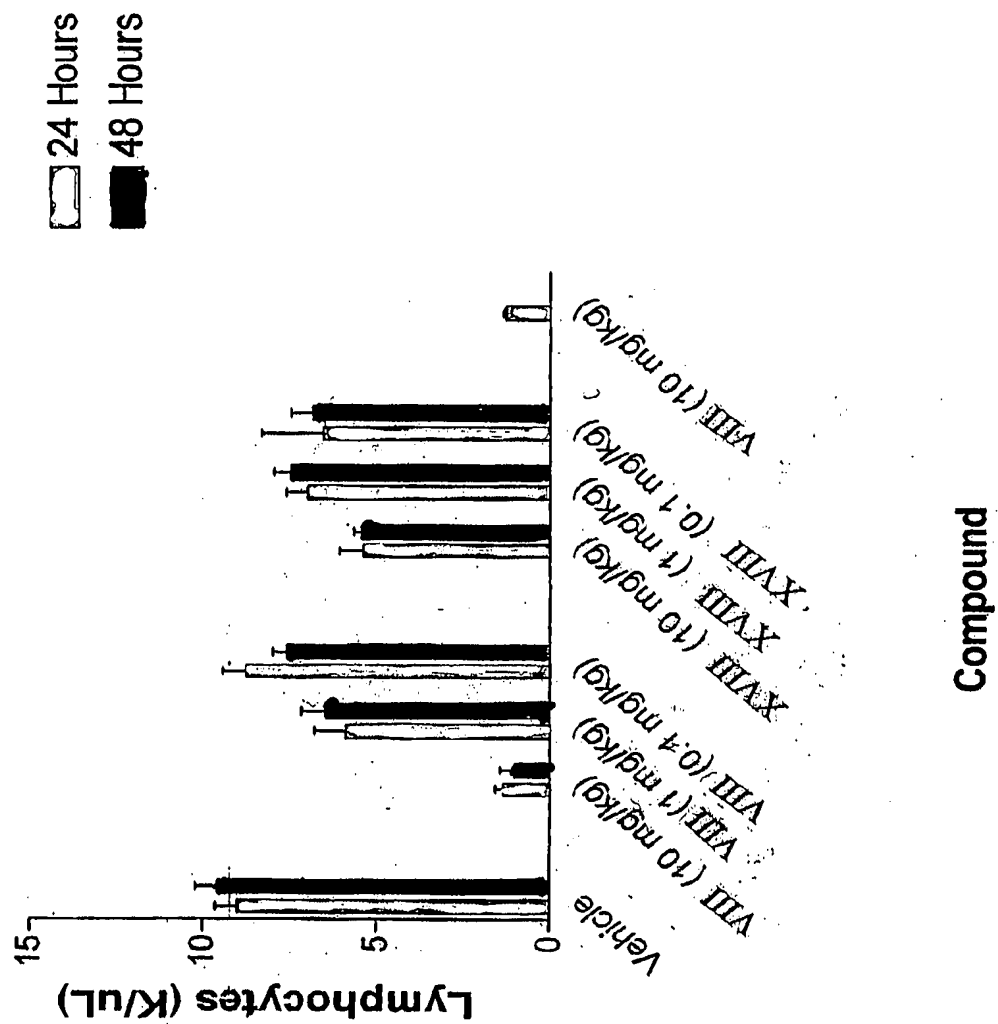


Fig. 10

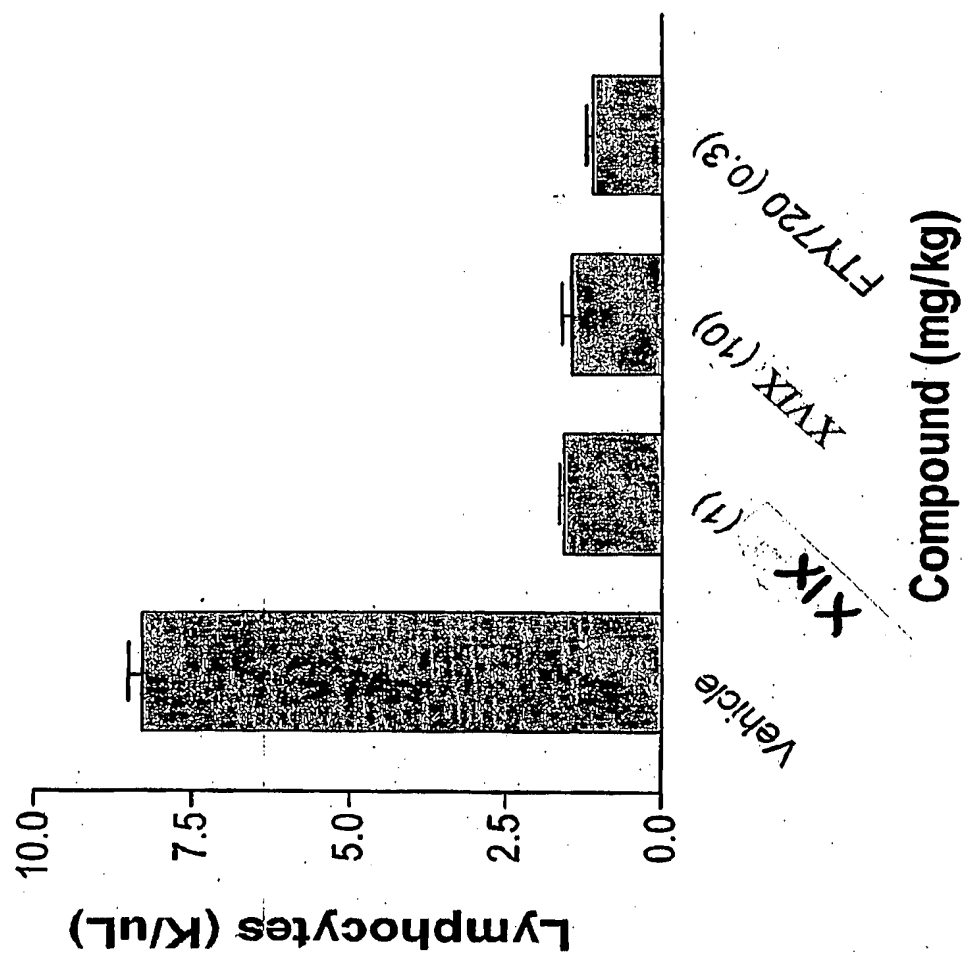
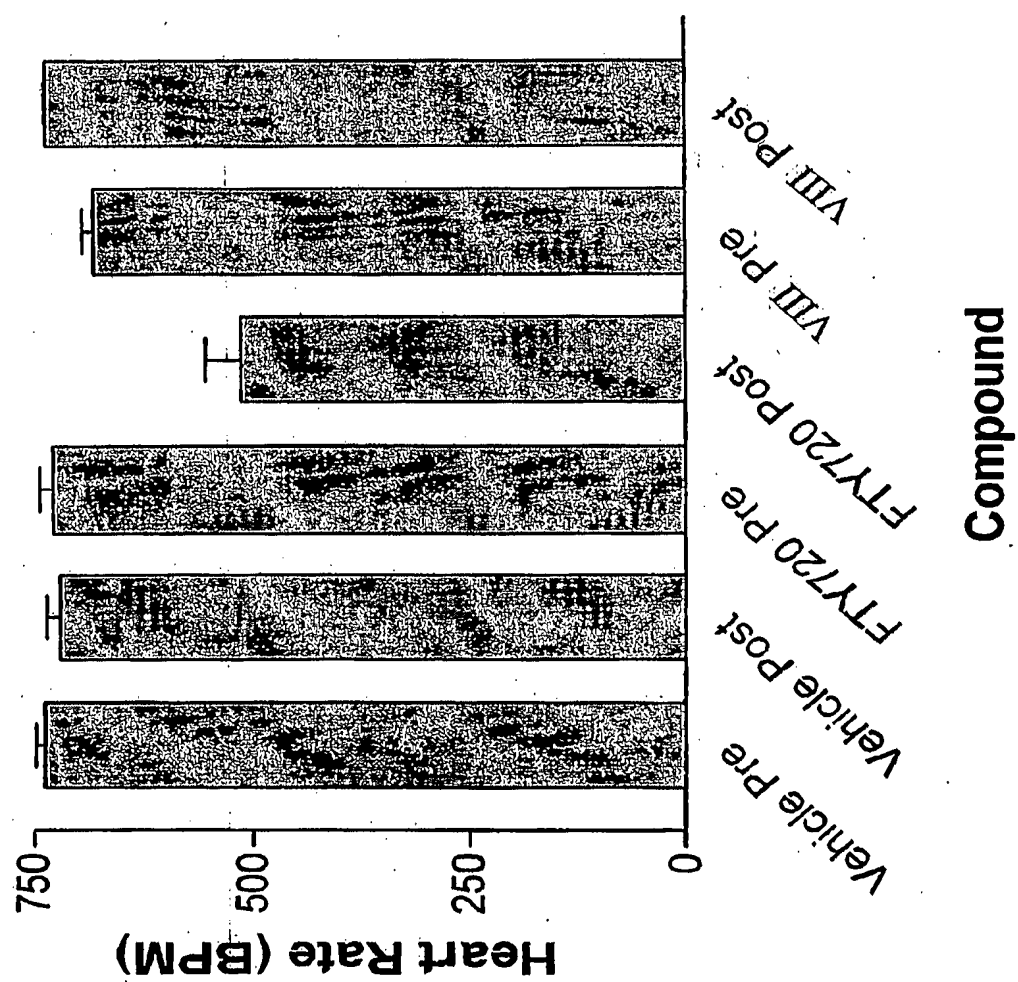


Fig. 11

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2007/003645

A. CLASSIFICATION OF SUBJECT MATTER INV. C07D271/06 C07D333/16 C07D419/04 C07F9/09 C07C215/08 C07C215/10 A61P29/00 A61P3/00 A61P19/00 A61K31/133 A61K31/381 A61K31/4245 A61K31/661 A61K31/6615					
According to International Patent Classification (IPC) or to both national classification and IPC					
B. FIELDS SEARCHED Minimum documentation searched (classification system followed by classification symbols) C07D C07F C07C A61K A61P					
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched					
Electronic data base consulted during the international search (name of data base and, where practical, search terms used) EPO-Internal, CHEM ABS Data, WPI Data, BEILSTEIN Data					
C. DOCUMENTS CONSIDERED TO BE RELEVANT					
Category*	Citation of document, with indication, where appropriate, of the relevant passages				Relevant to claim No.
X	BANDINI, MARCO ET AL: "An effective and useful synthesis of enantiomerically enriched arylglycinols" EUROPEAN JOURNAL OF ORGANIC CHEMISTRY , (10), 1937-1942 CODEN: EJOCFK; ISSN: 1434-193X, 2001, XP002440437 compound (8)				1-3, 17-19
X	KAWASAKI K-I ET AL: "Enantioselective Allylic Oxidation of Cycloalkenes by Using Cu(II)-Tris(oxazoline) Complex as a Catalyst" TETRAHEDRON, ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, NL, vol. 53, no. 18, 5 May 1997 (1997-05-05), pages 6337-6350, XP004105633 ISSN: 0040-4020 compound 2G				1-3, 17-19
-/--					
<input checked="" type="checkbox"/> Further documents are listed in the continuation of Box C. <input checked="" type="checkbox"/> See patent family annex.					
* Special categories of cited documents : <div style="display: flex; justify-content: space-between;"> <div style="width: 45%;"> <p>*A* document defining the general state of the art which is not considered to be of particular relevance</p> <p>*E* earlier document but published on or after the international filing date</p> <p>*L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>*O* document referring to an oral disclosure, use, exhibition or other means</p> <p>*P* document published prior to the international filing date but later than the priority date claimed</p> </div> <div style="width: 45%;"> <p>*T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>*X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone</p> <p>*Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.</p> <p>*G* document member of the same patent family</p> </div> </div>					
Date of the actual completion of the international search <div style="text-align: center; font-weight: bold;">3 July 2007</div>			Date of mailing of the international search report <div style="text-align: center; font-weight: bold;">16/07/2007</div>		
Name and mailing address of the ISA/ European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016			Authorized officer <div style="text-align: center; font-weight: bold;">Eberhard, Michael</div>		

INTERNATIONAL SEARCH REPORT

International application No

PCT/US2007/003645

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>CROSIGNANI S ET AL: "4-Naphthyl-Substituted Bis(Oxazoline): a New, Easily Recoverable and Efficient Chiral Ligand in Asymmetric Catalysis of the Diels-Alder Reaction" TETRAHEDRON, ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, NL, vol. 54, no. 51, 17 December 1998 (1998-12-17), pages 15721-15730, XP004144149 ISSN: 0040-4020 compounds (S)-8</p> <p>-----</p>	1-3, 17-19
X	<p>WO 2004/047743 A (SQUIBB BRISTOL MYERS CO [US]) 10 June 2004 (2004-06-10) example 92</p> <p>-----</p>	1-3, 17-19
A	<p>WO 2005/118523 A (NOVARTIS AG [CH]; NOVARTIS PHARMA GMBH [AT]; HINTERDING KLAUS [DE]; HO) 15 December 2005 (2005-12-15) page 18, line 20 - page 20, line 17 examples 11,12,20</p> <p>-----</p>	1-33
A	<p>WO 2004/096757 A1 (NOVARTIS AG [CH]; NOVARTIS PHARMA GMBH [AT]; ALBERT RAINER [CH]; EHRHA) 11 November 2004 (2004-11-11) abstract; examples 1-50</p> <p>-----</p>	1-33

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2007/003645

Box II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 22-29 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.
2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/US2007/003645

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2004047743 A	10-06-2004	AU 2003294441 A1 EP 1581510 A2	18-06-2004 05-10-2005
WO 2005118523 A	15-12-2005	AU 2005250112 A1 CA 2562332 A1 CN 1946679 A EP 1753712 A1 KR 20070015439 A	15-12-2005 15-12-2005 11-04-2007 21-02-2007 02-02-2007
WO 2004096757 A1	11-11-2004	AU 2004234067 A1 BR PI0409977 A CA 2523677 A1 EP 1622866 A1 JP 2006524662 T MX PA05011597 A US 2006211656 A1	11-11-2004 09-05-2006 11-11-2004 08-02-2006 02-11-2006 15-12-2005 21-09-2006